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THE ANTERIOR LOBE OF THE PITUITARY BODY IN ITS RELATIONSHIP TO THE EARLY GROWTH PERIOD OF BIRDS

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PATHOLOGICAL conditions in the pituitary body have been so frequently connected with abnormal growth that the former are generally conceded to bear a causal relation to the latter. Nevertheless attempts to modify growth by increasing the normal amount of pituitary secretion present in the body have yielded various and somewhat ill-defined results.

Caselli[1] noted no effect on growth after long-continued injections of whole pituitary glycerine extracts but found that ingestion retarded growth in some instances. Foderà and Pittau[2] observed that emaciation resulted from injection of pituitary extracts. Sandri[3] fed large quantities of ox pituitary to young mice for a period of two months and found that there was a notable arrest of growth. He also injected guinea pigs with a pituitary emulsion and found again a diminution in the rate of growth. He fails to state that he used control animals. Crowe, Cushing, and Homans,[4] using boiled suspensions of powdered pituitary from either dog, pig, or ox, found that repeated injections of the entire body caused rapid loss in weight both in puppies and in adult dogs. Pure anterior lobe preparations had no such

[1] Caselli: Revista sperimentale di freniatria. Reggio-Emilia, 1900 xxvi, pp. 176, 486.

[2] Foderà and Pittau: Gazzetta di medicina e chirurgia, 1909, viii, p. 149.

[3] Sandri: Archives italiennes de biologie, 1909, li, p. 337.

[4] Crowe, Cushing and Homans: Bulletin of the Johns Hopkins Hospital, 1910, xxi, p. 127.

effect, a normal puppy being given daily injections for three months without visible result. Also, according to Cushing,[1] Goetsch and Cushing found that one puppy fed daily with three grain doses of powdered extract of whole pituitary was less in height at the end of the experiment than the control.

Cerletti[2] worked upon guinea pigs and rabbits, using whole pituitaries of young sheep from which he prepared a glycerine emulsion. He injected an amount about equal to three fourths of a whole pituitary into the peritoneal cavity every five or six days. He also used dogs, giving them a more frequent dosage. All of his experiments covered considerable periods, the longest being 144 days. His results were uniform throughout. During the period of the experiment the animals receiving injection of pituitary extract fell constantly below the controls in weight. Moreover in the case of the dogs, measurement of the hind legs showed that those of the control were increasingly greater in length throughout the experiment than those of the dogs receiving pituitary injection. Measurement of the tibia in two experiments with rabbits showed that the diaphyses of the control animals were longer, but that the frontal diameter of the epiphyses of the animals receiving pituitary injection was equal to or greater than that of the controls.

Schäfer[3] fed young rats on a constant small amount of dried anterior lobe added to a measured diet of bread and milk. Fresh ox pituitaries were kept for a few days in chloroform, were then separated into anterior and posterior lobes, finely divided and dried. The controls were fed upon similarly prepared ovary or testicle. A group of four rats was fed in this way for three months. For the first eight days the pituitary fed rats fell below the controls in weight, but at the close of the experiment their total weight was almost twice that of the controls. In a second series of experiments similarly conducted, Schäfer[4] arranged the animals in three groups, the first containing four young females, the second three young males, the third three half grown males,

[1] CUSHING: The pituitary body and its disorders, 1912.
[2] CERLETTI: Archives italiennes de biologie, 1907, xlvii, p. 123.
[3] SCHÄFER: Proceedings of the Royal Society, 1909, lxxxi, B, p. 442.
[4] SCHÄFER: Quarterly journal of experimental physiology, 1912, v, p. 203.

each group being duplicated by a control group. The experiment continued for three months. At first ¾ gm. of material was administered. This amount was doubled after two months and still further increased toward the end of the experiment. At the conclusion the ovary fed animals in the first two groups were a little heavier than the pituitary fed animals. In the third group the pituitary animals about equaled the ovary fed animals in weight. The conclusion is drawn from these experiments that the addition of small amounts of ovarian or pituitary tissue to the diet of rats has little or no effect upon growth.

Aldrich[1] performed two series of experiments, the first upon dogs, the second upon rats. He added to the bread and milk diet of each about 50 mg. per day of fresh, dessicated, defatted anterior lobe of ox pituitary. He used seven dogs divided into two groups, one group being fed upon similarly prepared ovary as control. The average weights show that the controls had a natural tendency to increase in weight more rapidly than the pituitary animals, but for certain reasons Aldrich concludes that there is neither stimulation nor retardation in growth, though in individual cases the anterior lobe may stimulate. In the second series, Aldrich[2] used ten young rats divided into two groups, male and female in each. The control group, fed as in the case of dogs, held an increasingly greater weight than the pituitary group during the three months of the experiment. Ingestion of the anterior lobe thus impedes growth. A similar series performed with posterior lobe gave no such inhibition. Aldrich mentions that J. L. Miller has conducted experiments on young white rats in much the same way and has obtained negative results as regards weight and skeletal change.

The preponderance of the above evidence indicates that the pituitary body either injected or ingested is able to cause a diminution in rate of growth in young animals. That the falling off in weight is due to something more than emaciation has been shown by those investigators who have through measurement of the long bones found a decrease in their length. I have been

[1] ALDRICH: This journal, 1912, xxx, p. 352.
[2] ALDRICH: This journal, 1913, xxxi, p. 94.

able to confirm in this series of experiments both the falling off in weight of the animal and in length of bone following ingestion of anterior lobe of the pituitary body.

Experimental

In order that dependable averages might be secured through the use of a number of individuals of the same age, the domestic fowl was selected for the investigation. The work was started September 25 with two groups each containing eighteen White Leghorn chicks either two or nine days old at that time. Feeding with pituitary material began October 3 and continued until January. The chicks were kept within a small enclosure in the laboratory and given artificial heat until they reached 250 gm. in weight when they were allowed the range of a sunny room. They were fed at first upon finely cracked grains with a little boiled rice, then upon coarsely cracked grains, entire wheat, grit, and a mash composed of bran, shorts, bone meal, cornmeal and charcoal. They were also given green feed. Five days in the week each chick was fed a weighed amount of unmodified anterior lobe of ox pituitary or in the control group an equal amount of fresh liver. The ox pituitaries were obtained from the Oakland Meat and Packing Company through the great courtesy of the Superintendent. The glands were eaten by the chicks within twenty-four hours after being removed from the cattle at the slaughter house. The amount of pituitary material given varies throughout the experiment, the attempt being to keep it roughly equal to one hundredth of the average body weight of the pituitary chicks. A few individuals died early in the experiment. During December probably on account of the confinement and adverse weather conditions, the chickens contracted roup. On this account it was thought best to give figures for two months only and to gather the data from those individuals which up to December appeared normal. If all had been used the result would have been to reduce the averages of the pituitary fed individuals still further, as the very smallest individuals in the pituitary group were eliminated in this way, while those eliminated from the controls were of fairly large size. It should also be observed that

the results obtained during the first two months were continued without change during the third month. The extension of the curves in the direction in which they are going would indicate in a general way the growth occurring in this last interval.

The results here presented are based upon data gathered from twenty-five individuals divided into four groups: (1) 7 pituitary fed females, (2) 10 liver fed females, (3) 4 pituitary fed

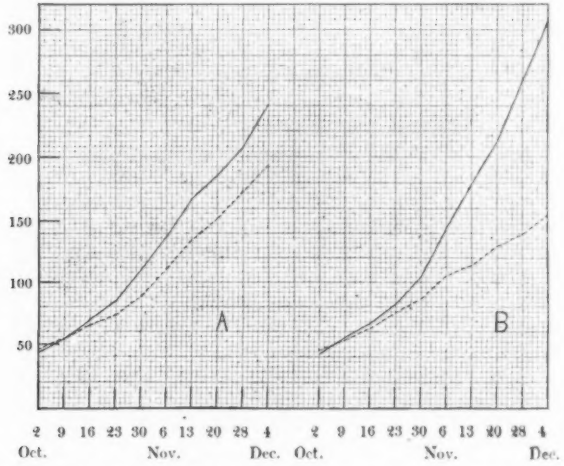


FIGURE 1. Derived from Table I. Abscissae: successive dates. Ordinates: weight in grams. Broken lines: data from pituitary fed chickens. Solid lines: data from controls. The curves represent the average weight in successive weeks of (A) 7 pituitary fed females and 10 control females, (B) 4 pituitary fed males and 4 control males.

males, (4) 4 liver fed males. Measurements were taken at weekly intervals. Table I, represented graphically by the accompanying curves (Figs. 1 and 2), gives for each group (1) the average weight in grams, (2) the average length of wing in millimeters, (3) the average length of foot in millimeters. The length of wing was measured by placing the joint between radius and metacarpus against an upright and measuring on a horizontal meter stick to the tip of the feathers. The foot was measured in a similar manner from the joint between tibio-tarsus and tarso-metatarsus to the end of the central toe.

All the measurements show a distinct inhibition of growth in

the pituitary fed chickens both male and female. It is noticeable that the males are more affected than the females. At the conclusion of the experiment, while the average measurements of the male controls were greater than those of the female controls, the average measurements of the pituitary fed males were less than those of the pituitary fed females. The measurements of all

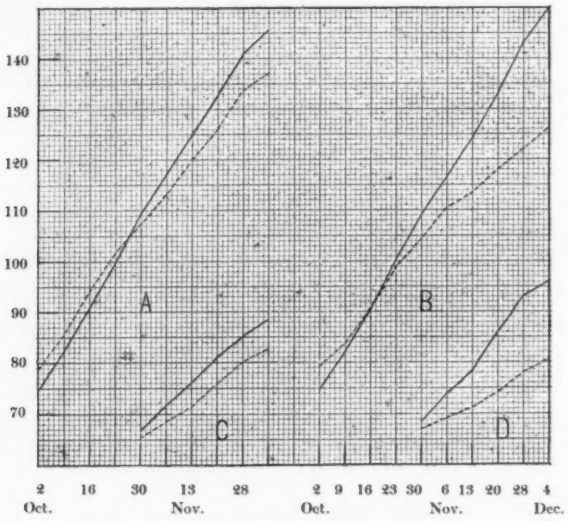


FIGURE 2. Derived from Table I. Abscissae: successive dates. Ordinates: length in millimeters. Broken lines: data from pituitary fed chickens. Solid lines: data from controls. The curves represent the average length in successive weeks of (A) the wings of 7 pituitary fed females and 10 control females, (B) the wings of 4 pituitary fed males and 4 control males, (C) the feet of 7 pituitary fed females and 10 control females, (D) the feet of 4 pituitary fed males and 4 control males.

the individuals included in the totals for December 4 present this fact in another way (Table II). It will be observed that whereas the two groups of females overlap one another in weight and length of bone, the male groups show no overlapping in any measurement. This difference could hardly be entirely due to the smaller number of males.

The inhibition of growth indicated in the tables was easily apparent on inspecting the living animals. Those fed upon

TABLE I

Date	Grams Pituitary or Liver fed	Female						Male					
		Pituitary fed Average of 7			Control Average of 10			Pituitary fed Average of 4			Control Average of 4		
		Wt. gm.	Wing mm.	Foot mm.	Wt. gm.	Wing mm.	Foot mm.	Wt. gm.	Wing mm.	Foot mm.	Wt. gm.	Wing mm.	Foot mm.
Oct. 2	.3	45.4	78.7		43.5	74.8		45.0	79.2		41.5	74.7	
" 9	.4	54.1	85.8		53.9	82.4		53.0	83.7		55.5	82.2	
" 16	.6	65.4	94.2		69.6	90.9		62.7	90.7		65.7	90.5	
" 23	.8	73.2	101.7		84.5	99.8		75.2	99.0		81.5	100.5	
" 30	.9	88.7	107.4	65.4	109.9	109.3	66.9	86.0	104.5	67.0	104.0	109.0	68.5
Nov. 6	1.1	111.7	113.2	68.5	136.9	117.2	71.7	105.2	110.5	69.2	144.0	116.5	74.0
" 13	1.3	133.8	120.0	71.4	167.3	124.8	76.1	113.2	113.5	71.2	178.5	124.2	78.2
" 20	1.3	151.8	126.2	76.0	185.7	132.9	81.0	129.0	118.0	74.2	211.5	133.2	86.0
" 28	1.4	173.2	133.7	80.1	207.3	140.9	85.1	139.5	122.2	78.0	261.0	143.2	93.2
Dec. 4	1.5	192.8	137.1	82.7	240.3	145.5	88.5	154.0	126.2	80.5	308.0	150.5	96.2

pituitary material were noticeably smaller, this difference being especially prominent in the males. After three months, the three smallest males, all pituitary fed, showed no wattles and

FIGURE 3. Photograph taken November 29. The posterior chicken is a control male. The anterior one is a pituitary fed female. Note how the control exceeds in length of foot, wing and tail and in height. The female could have been exchanged for a male in the same group without altering the appearance of the picture.

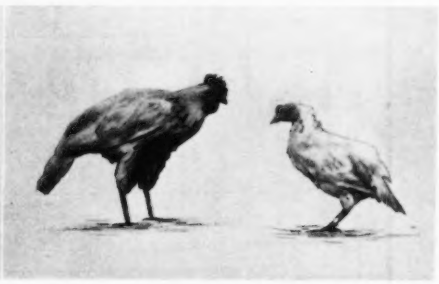


FIGURE 4. Photograph taken about January 15. The larger chicken is a control male, the smaller a pituitary fed male. Besides differences in size of all parts of the body, note that the pituitary fed chicken has no wattles and a very small comb as compared with the well developed comb and wattles of the control.

their combs were only slightly larger than those of the females, whereas the combs and wattles of the three remaining control males were large and well developed. (Figs. 3 and 4.)

These results are reinforced by a previous feeding experiment in which three White Leghorn fowls were raised to adult size. One, the control, was a male. The others, fed as above on pituitary material, were male and female. This male originally exceeded the control in size, but in the course of a few days it dropped below and remained smaller than the control during the period of growth corresponding to that covered by the tables given above. The female was always the smallest of the three.

Involution of the thymus. After two months' feeding, the smallest of the pituitary fed chickens died. Autopsy showed that there was practically no thymus tissue left. For comparison the smallest of the controls was killed. It was found to have a considerable amount of thymus tissue stretching through the neck in close contact with both jugular veins. This led to an

TABLE II

Female Pituitary fed			Female Control			Male Pituitary fed			Male Control		
Wt. gm.	Wing mm.	Foot mm.	Wt. gm.	Wing mm.	Foot mm.	Wt. gm.	Wing mm.	Foot mm.	Wt. gm.	Wing mm.	Foot mm.
129	128	73				124	134	85			
134	125	72				156	118	80			
			162	133	79	157	123	76			
			185	137	84	179	130	81			
194	135	87							206	142	95
196	140	85							322	148	96
			202	134	83				347	160	95
			218	139	88				357	152	99
			222	139	86						
231	142	85									
			232	150	81						
238	150	87									
			254	155	92						
			279	157	96						
			320	163	99						
			329	148	97						

examination of the thymus in the living chickens. Long areas of skin free from feathers extend along the dorsal aspect of the chicken's neck, and by parting the feathers to lay these bare and also stretching and moving the skin it is possible to see both jugular veins quite plainly with their attendant masses of thymus tissue. Although some of the thymus which lies close to the thorax may be overlooked, a good idea of the amount of thymus tissue may be gained in this way. By means of the examination, which was made in the early part of December, it was found

possible to arrange the chickens in a somewhat accurate series from the one possessing the smallest to the one possessing the largest thymus. This agreed in general with the order of arrangement by weight including both males and females. Table III shows the distribution of the pituitary and the control chickens according to the size of the thymus. On weighing the total

TABLE III

Group I Thymus small	Group II Thymus somewhat larger	Group III Thymus medium	Group IV Thymus large
7 pituitary 3 males 4 females	3 pituitary 1 male 2 females	1 pituitary female	
1 control female	3 controls 1 male 2 females	4 controls all females	6 controls 3 males 3 females

thymus tissue in representatives of Groups I, II, and IV, it was found that the specimen thymus from Group I (body wt. 148 gm., thymus wt. .1038 gm.) was .07% of the body weight. In group II, it was .14% of the body weight (body wt. 229 gm., thymus wt. .321 gm.). In Group IV it was .31% of the body weight (body wt. 299 gm., thymus wt. .953 gm.). Thus there is more than four times as much thymus tissue per gram of body weight in the representative of Group IV as in the representative of Group I. It will be noticed that Group IV is made up entirely of control animals whereas all the small thymuses except one belong to pituitary fed animals. Here again the males will be seen to be more widely separated than the females, all the male pituitary chickens having small thymuses whereas all the male controls had very large thymuses except the one in Group II which soon after became sick.

Although I can find in the literature no observations concerning the involution of the thymus under pituitary feeding, the converse condition has been observed. Aschner[1] notes that the

[1] ASCHNER: Archiv für die gesammte Physiologie, 1912, cxlvi, p. 1.

thymus persists abnormally in young dogs from which the pituitary has been removed, which is in accordance with the retention of certain infantile characteristics. The same observation has been made by Cushing,[1] who further adds that clinical experience

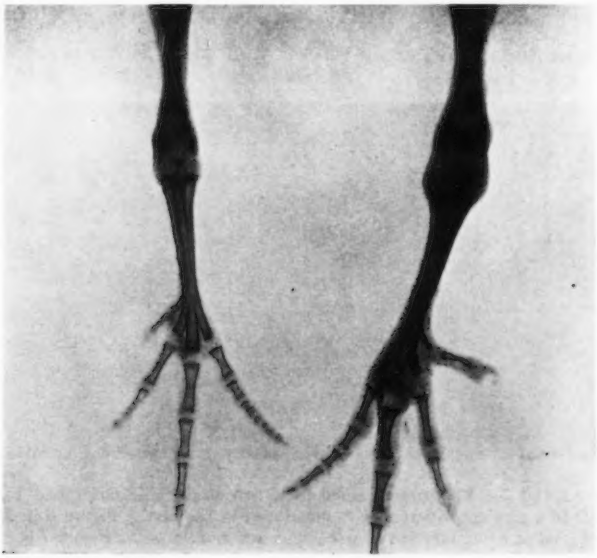


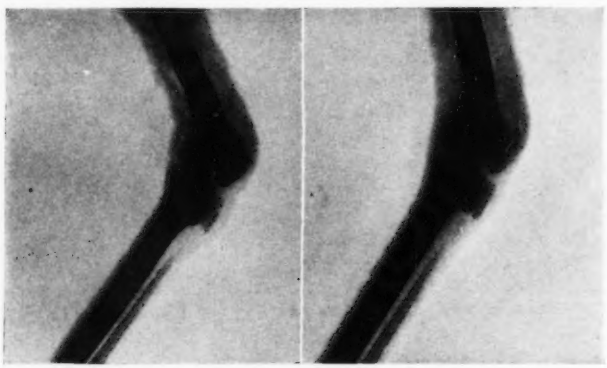
FIGURE 5. Radiogram taken December 5. The larger leg is that of a control female, the smaller that of a pituitary fed male. At the time each was the closest representative of the average in its own group. Note the greater length of bone in the foot and the larger and more plainly defined centers of ossification at the tarsal joint in the control.

would lead him to believe that in cases of hypophyseal insufficiency there is apt to be a persistent and enlarged thymus where the process dates from a preadolescent period.

It is possible that the diminution in growth which accompanies injection and ingestion of pituitary tissue may be caused indirectly by disturbances in nutrition. But it is also possible that this inhibition may be the result of a primary effect of pituitary secretion upon the thymus. In fact the changes brought about through

[1] CUSHING: *Loc. cit.*

its action call to mind the changes found by Basch[1] to follow extirpation of the thymus in young dogs. In the operated animals the bones were slighter and smaller than in the controls while the epiphyseal lines were frequently broadened and irregular. There is a larger proportion of cartilaginous tissue at the epiphyseal line than in the normal animals. The cartilaginous covering of the epiphyses is thicker and more voluminous and the



FIGURES 6 and 7. Radiograms taken from two of the chickens raised to adult size in a previous experiment as mentioned in the text. Figure 6 is the tarsal joint of a pituitary fed female about five months old. Figure 7 is the tarsal joint of a control male of the same age. Compare especially the distal ends of the tibio-tarsus. That of the pituitary fed hen shows considerably more cartilage and less bone than that of the control, suggesting the changes in bony growth found by Basch to follow extirpation of the thymus.

ossified bony portion is smaller. The long bones take longer to develop and this causes a marked retardation in growth of the whole body. These words might be used in large part as a description of the differences existing between the bones of pituitary fed chickens and controls as shown by the accompanying radiograms. (Figs. 5, 6, and 7.) Whether or not the above idea is fruitful will require further experimentation to decide, and it is my purpose to continue the study.

[1] BASCH: Jahrbücher für Kinderheilkunde, 1906, lxiv.

Conclusions

1. The growth of young fowl is retarded by the addition to the diet of fresh, unmodified anterior lobe of ox pituitary. This is shown both in body weight and in length of the long bones.

2. Involution of the thymus accompanies this retardation and may bear a causal relation to it.

3. These effects are more marked in the males than in the females.

The writer extends grateful thanks to Professor Maxwell for his constant and kindly suggestions and help.

THE RÔLE OF NASCENT OXYGEN IN REGULATING THE ACTIVITIES OF ENZYMES IN ANIMALS AND PLANTS

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IT may be shown in several ways that nascent oxygen destroys all the ordinary enzymes.

We have found that ptyalin, amylopsin, malt diastase, rennin, pepsin and trypsin are destroyed by the passage of the direct electric current and that the rate of this destruction is directly proportional to the amount of current used. Since the time of Faraday it has been known that the passage of the direct electric current decomposes water and that the amounts of oxygen and hydrogen liberated are directly proportional to the amount of current passed. We have no evidence that nascent hydrogen decreases the activity of enzymes, but we have abundant evidence that nascent oxygen decreases their activity and that this decrease is more or less proportional to the amount of oxygen used. Hence it seems reasonable to conclude that the cause of the destruction of enzymes by the passage of the direct electric current is oxidation.[1]

The above named enzymes are also destroyed by bubbling oxygen gas or air through their solutions, but the rate of this destruction is very slow. However, if a piece of platinum mesh, previously covered with platinum black, be introduced into the solution, thereby supplying nascent oxygen, the rate of the destruction of the enzyme is greatly increased.[1]

We have been able to show in still another way that nascent oxygen destroys the activity of enzymes. If hydrogen peroxide be added to the enzyme solution and a piece of platinum mesh,

[1] Burge: Résumés des Communications, IXième Congrès international des Physiologistes, Groningue. 1913.

previously covered with platinum black, be introduced the oxygen liberated from the hydrogen peroxide by the platinum black oxidizes the enzyme. This method was used in the experiments to be described in this paper.

The solutions were made by dissolving one gram of a commercial preparation of the enzyme in 15 cubic centimeters of distilled water. Such solutions possessed optimum activity. Two cubic centimeters of the enzyme solution were used in each experiment. This amount was diluted to 7 cubic centimeters using varying amounts of hydrogen peroxide and a diluting solution. The diluting solution consisted of some of this solution of hydrogen peroxide which had been decomposed completely by means of platinum black. The strengths of the diastatic enzymes were determined by the amounts of reducing sugar produced by the addition of 2 cubic centimeters of the solution to 10 cubic centimeters of a 0.2% starch paste. The digestion was carried on in the case of amylopsin and ptyalin for three minutes at 38° C. In the case of malt diastase and taka diastase the amounts of starch paste and of the enzyme solution were the same as for amylopsin and ptyalin, but the digestion was carried on for thirty minutes at

TABLE I

Ezyme	N	A	B	C	D	E
	2 c.c. enzyme 5 c.c. diluting solution 0 c.c. H_2O_2	2 c.c. enzyme 4 c.c. diluting solution 1 c.c. H_2O_2	2 c.c. enzyme 3 c.c. diluting solution 2 c.c. H_2O_2	2 c.c. enzyme 2 c.c. diluting solution 3 c.c. H_2O_2	2 c.c. enzyme 1 c.c. diluting solution 4 c.c. H_2O_2	2 c.c. enzyme 0 c.c. diluting solution 5 c.c. H_2O_2
Amylopsin	9.8 mgs.	6.8 mgs.	5.0 mgs.	2.6 mgs.	0.0 mgs.	0.0 mgs.
Ptyalin	7.5 "	7.3 "	7.5 "	7.2 "	2.4 "	0.0 "
Malt Diastase	9.4 "	5.4 "	4.2 "	2.8 "	0.0 "	0.0 "
Taka Diastase	8.9 "	8.4 "	8.6 "	8.9 "	8.7 "	8.8 "
Pepsin	1.4 mm.	1.2 mm.	1.2 mm.	1.0 mm.	0.8 mm.	0.2 mm.
Rennin	30″	50″	60″	75″	540″	8 hrs.

60° C. Pavy's method was used for the estimation of the sugar. Mett's tubes were used in determining the strength of the pepsin solutions. The activity of the rennin was determined by adding 1 cubic centimeter of the solution to 5 cubic centimeters of fresh cow's milk at 38° C.

The details of these experiments may be seen in the accompanying table.

The solutions indicated in column N were made by adding 5 cubic centimeters of the diluting solution to 2 cubic centimeters of the enzyme solution of optimum activity. The solutions in column A were made by adding 4 cubic centimeters of the diluting solution and 1 cubic centimeter of hydrogen peroxide to 2 cubic centimeters of the concentrated enzyme solution. Solutions in column B were the same as those in column A except that 3 cubic centimeters of the diluting solution and 2 cubic centimeters of hydrogen peroxide were used. The solutions indicated in columns C, D and E were similar to those in A and B except for the increasing amounts of hydrogen peroxide and the decreasing amounts of diluting solution as shown in the table.

The experiments were made as follows: A piece of platinum mesh, previously covered with platinum black by the passage of the direct electric current, was thoroughly washed and introduced into a solution of amylopsin made by adding 5 cubic centimeters of the diluting solution to 2 cubic centimeters of the concentrated amylopsin solution (column N). This was allowed to stand for thirty minutes. It will be noticed that no oxygen was given off to this solution. Two cubic centimeters of this solution were added to 10 cubic centimeters of a .2% starch paste and permitted to stand at 38° C. for three minutes. At the end of this time the digestion was brought to a close by bringing the solution to boiling in thirty seconds. It may be seen in column N for amylopsin that 9.8 milligrams of reducing sugar were present as determined by Pavy's method. The same piece of platinum mesh was introduced into another solution of amylopsin made by adding 4 cubic centimeters of the diluting solution and 1 cubic centimeter of hydrogen peroxide to 2 cubic centimeters of the concentrated solution of amylopsin. This solution was permitted to stand until the 1 cubic centimeter of hydrogen peroxide was completely de-

composed by the platinum mesh. The diastatic power of this solution was then determined as before and in column A it may be seen that this was represented by 6.8 milligrams of reducing sugar. In column B for amylopsin may be seen the result of a similar experiment except that 3 cubic centimeters of hydrogen peroxide were used. The diastatic power of the amylopsin was here represented by 5.0 milligrams of reducing sugar. The experiments in column C, D and E for amylopsin were made in a similar way with the solutions indicated in the table. In each of these solutions was placed a piece of platinum mesh and each was allowed to stand until all the hydrogen peroxide was decomposed. In column C for amylopsin the diastatic power of the solution is represented by 2.6 milligrams of reducing sugar, and in column D the diastatic power is reduced to O. In column C the solution was exposed to the oxygen liberated from the decomposition of 3 cubic centimeters of hydrogen peroxide. In column D the solution was exposed to the oxygen liberated from 4 cubic centimeters of hydrogen peroxide. This amount was sufficient to oxidize completely the amylopsin present.

It may be seen that the normal diastatic power of the amylopsin used in this experiment is represented by 9.8 milligrams of reducing sugar, and that when a similar amount of amylopsin was exposed to the action of the oxygen liberated from 1 cubic centimeter of hydrogen peroxide its diastatic power was reduced to 6.8 milligrams of reducing sugar, i.e., the amount of oxygen that can be liberated from 1 cubic centimeter of hydrogen peroxide reduced the diastatic power of the amylopsin by 3.0 milligrams of reducing sugar. It will be observed for amylopsin in column B that the oxygen liberated from 2 cubic centimeters of hydrogen peroxide reduced the diastatic power by 4.8 milligrams of reducing sugar, or 2.4 milligrams of sugar per cubic centimeter of hydrogen peroxide. In column C the oxygen from 3 cubic centimeters of hydrogen peroxide reduced the diastatic power by 7.2 milligrams of reducing sugar, or 2.5 milligrams per cubic centimeter of hydrogen peroxide. In column D the oxygen liberated from 4 cubic centimeters of hydrogen peroxide completely destroyed the dastatic power of the amylopsin.

Similar experiments to those on amylposin were carried out on

ptyalin, malt diastase, taka diastase, pepsin and rennin. It may be seen for ptyalin in column N that when 2 cubic centimeters of the concentrated solution of ptyalin were diluted to 7 cubic centimeters by adding 5 cubic centimeters of the diluting solution the diastatic power is represented by 7.5 miligrams of reducing sugar; that when a similar amount of a solution of ptyalin was exposed to the action of the amount of oxygen liberated from 1 cubic centimeter of hydrogen peroxide the diastatic power is represented by 7.3 milligrams of reducing sugar and that when it was exposed to the amount of oxygen liberated from 5 cubic centimeters of hydrogen peroxide by platinum black its diastatic power was reduced to 0. It will be noticed that the rate of the destruction of ptyalin was not proportional to the amount of hydrogen peroxide added and hence not to the amount of oxygen liberated. It may be seen also that a similar destructive action is produced on malt diastase by the oxygen liberated from hydrogen peroxide. Its diastatic power was completely destroyed when its solution was exposed to the amount of oxygen liberated from 4 cubic centimeters of hydrogen peroxide. The solutions of taka diastase were not affected by the amounts of oxygen used in these experiments. However, the activity of taka diastase was destroyed by exposure to the amount of oxygen liberated from 12 cubic centimeters of hydrogen peroxide. The activity of both pepsin and rennin was greatly reduced but was not completely destroyed by the amount of oxygen obtained from 5 cubic centimeters of hydrogen peroxide. Both of these enzymes were destroyed by the oxygen liberated from 7 cubic centimeters of this hydrogen peroxide. The rates of the destruction of these two enzymes run parallel. This may be accounted for by the fact that the solutions of pepsin and rennin were both made from the same commercial preparation of pepsin. We do not think that the result is any proof for the identity of the two substances because if a commercial preparation be chosen which has strong rennetic property and weak peptic property it is possible to destroy the peptic property without apparently affecting the rennetic.

In addition to the enzymes enumerated in the table we have found that oxygen liberated from hydrogen peroxide by platinum black destroys the diastase extracted from Elodea canadensis

gigantea, trypsin, emulsin, invertase, bromalin, papain and autolytic enzymes.

Brown and Morris[1] have shown that the amount of diastase in the leaves of foliage plants increases during the night and decreases during the day. Using Elodea canadensis gigantea, a green water plant, we have been able to confirm their observations. We also found that the amount of destruction of the diastase was more or less proportional to the length of time of exposure of the plant to light. The fact that diastatic enzymes are destroyed by nascent oxygen would seem to offer an explanation of this observation that the diastase in plants decreases during the exposure of the plant to light. The assumption would be that the oxygen liberated during exposure to light oxidizes the diastase formed, hence a decrease in diastatic activity during the day. As soon as the plant is removed to darkness oxygen ceases to be given off and during this period the diastatic activity increases.

The oxidative action of the tissues has been demonstrated by various observers[2] employing a variety of reactions by which colored oxidation products are formed within the tissues. These observers[3] have shown that the capacity of the tissues to form these colored products bears a direct relation to their capability of freeing oxygen from hydrogen peroxide, of bluing tincture of guaiac and of oxidizing salicylic aldehyde and benzyl alcohol to their respective acids. They found that there are definite and constant differences in the oxidative properties of the different tissues. This property was most marked in the spleen, liver and kidney and least marked in muscular and nervous tissue.

Using the above methods Lillie[4] examined the oxidative properties of the different regions of the alimentary canal. When such sections were placed in a solution of alpha benzol and di-amidobenzene the parts of the cross section where oxidation takes places most rapidly were stained violet by the formation of indophenol dyes. He found that when cross sections of the stomach wall were placed in such solutions the mucous membrane assumed a

[1] Brown and Morris: Journal of the Chemical Society, 1893, 63, p. 604.
[2] Medwedew: Archiv für die gesammte Physiologie, 1896, 65, p. 249.
[3] Salkowski (mit Jamagiwa): Virchow's Archiv, 1897, 147, p. 1.
[4] Lillie: This journal, 1902, 7, p. 413.

deep violet coloration which was particularly intense at the inner ends of the cells. The muscular layers took on a diffuse and relatively slight coloration in accordance with the generally observed feeble oxidative properties of muscle. The connective tissue portions of the sub-mucosa remained almost colorless. Cross sections of regions of the intestine showed a similar distribution of the indophenol coloration. In all regions the mucosa colored soonest and most deeply while the muscular layer and the submucosa assumed a relatively slight stain. Thus he concludes that the mucosa in both stomach and intestine possesses intense oxidative properties.

The fact that pepsin and trypsin are easily oxidized and that the mucosa of the stomach and intestine possesses intense oxidative properties would seem to offer an explanation of the fact that these organs are not digested by the pepsin and trypsin contained within their lumen. The assumption would be that the pepsin and trypsin immediately in contact with the mucosa of the stomach and intestine respectively undergo oxidation and that by such means the cells maintain their integrity during life.

Salkowski and others [1] have shown that all the body tissues possess the power of undergoing autolysis after death and that under certain normal as well as pathological conditions tissues and even organs may undergo autolysis during life. The atrophy of the thymus and the involution of the puerpural uterus might be mentioned as examples of normal auto-digestion. Various theories have been advanced to account for the fact that the tissues do not undergo auto-digestion during life as after death. One theory [2] is that there are in the living tissues anti-substances which hold the autolytic enzymes in check. Another theory [3] suggests that the tissues are protected by their alkaline reaction as it has been shown that an acid reaction is necessary for the activity of autolytic enzymes. A third theory assumes that the enzymes exist in a zymogen form and are activated or inactivated as the need may arise.

In view of the fact that autolytic enzymes in common with all

[1] SALKOWSKI: Deutsche Klinik, 1903 (11), 147.
[2] GLAESSNER: Hofmeister's Beiträge, 1904, 4, 79.
[3] WIENER: Centralblatt für Physiologie, 1905, 19, 349.

the ordinary enzymes are destroyed by nascent oxygen an additional theory may be advanced, namely that the tissues maintain their integrity during life by means of their oxidative properties. This theory would assume that normally a balance exists between the autolytic enzymes and the oxidative processes of the tissues. It is known that in infectious diseases,[1] in diseases of the circulatory and respiratory systems,[2] in acute yellow atrophy of the liver and in chloroform and phosphorus poisoning autolysis may be increased to a marked degree. Without the oxygen continually supplied by the circulatory and respiratory systems oxidation in the tissues would be impossible. Since this is true, any special interference with either of these systems would presumably result in a decreased oxidation in the tissues. If the balance which has been assumed to exist between the oxidative and autolytic processes exists, then any interference with the supply of oxygen to the tissues should express itself in an increased rate of autolysis. Schlesinger [2] noted an intense self-digesting tendency of the tissues in diseases of the circulatory and respiratory systems. Under such conditions the amount of oxygen supplied to the tissues is decreased and the fact that under these conditions autolysis is increased would seem to support the above assumption. Jacoby [3] showed that the livers of dogs dead of phosphorus poisoning underwent autolysis more rapidly than normal livers, while Welsch [4] and Riess [5] found that oxidation in cases of this poisoning is decreased. Welsch made a study of the respiratory exchange in cases of phosphorus poisoning and found that the oxidative processes were decreased by about 20%. Riess proved the deficiency of oxidation also by showing the presence in the urine of large amounts of organic acids which are oxidized under normal conditions.

[1] Flexner: University of Pennsylvania Bulletin, July, 1903.

[2] Schlesinger: Hofmeister's Beiträge, 1904, 4, 87.

[3] Jacoby: Zeitschrift für physiologische Chemie, 1900, 30, 174.

[4] Welsch: Archives internationales de pharmacodinamie et de Thérapie, 1905, 14, 211.

[5] Riess: Berliner klinische Wochenschrift, 1905 (42), 44a, 54.

Conclusions

1. The facts that pepsin and trypsin are oxidized by nascent oxygen and that the mucosa of the stomach and intestine possesses intense oxidative properties may be used to explain the protection of these organs from self-digestion during life.

2. The fact that disatase is destroyed by nascent oxygen offers an explanation of the observation that the amount of this enzyme is decreased during the day and increased during the night in plants.

3. The fact that the autolytic enzymes are destroyed by nascent oxygen and that the tissues possess oxidative properties would seem to justify the assumption that normally there is a balance between the oxidative and autolytic processes in the living tissues. The fact that in certain pathological conditions where autolysis is increased the oxidative processes are decreased is in accord with this assumption.

CONTRIBUTIONS TO THE PHYSIOLOGY OF THE STOMACH

XIV. The Influence of Smoking and of Pressure on the Abdomen (Constriction of the Belt) on the Gastric Hunger Contractions

By A. J. CARLSON and J. H. LEWIS

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IT is generally held to be true that smoking shortly before a meal leads to depression of hunger and appetite. It is also a common belief that strong pressure on the abdomen ("tightening the belt") decreases or relieves the hunger sensation, at least temporarily. We are now in position to test the correctness of these beliefs by decisive experiments, at least as regards the influence of these measures on the objective hunger contractions and the subjective hunger sensations.

I. The Influence of Smoking on the Hunger Contractions and on the Hunger Sensations

Depression or inhibition of hunger by smoking is rendered probable by the fact that anything which stimulates the sensory nerve endings in the mouth and in the gastric mucosa inhibits the gastric hunger contractions in direct proportion to the intensity of the stimulation.[1] Smoking stimulates the nerve ending in the mouth in varying degrees according to the kind of tobacco used. Smoking frequently involves stimulation of nerve endings in the gastric mucosa owing to the swallowing of saliva containing nicotin, oils, tannic acid, and probably other irritating substances. Smoking may also act on the hunger mechanism in a third way, that is, through absorption of nicotin and other products of the combus-

[1] Carlson: This journal, 1913, xxxi, p. 212.

tion. This third possibility has not been investigated. It is well established, however, that even small quantities of nicotin in the blood leads to nausea and vomiting. Nausea and vomiting are accompanied by atony of the gastric fundus, which insures absence of hunger contractions and hunger sensations.

The effects of smoking on the gastric hunger contractions were first studied on Mr. V., our young man with the permanent gastric fistula. In his case smoking (cigars) leads invariably to inhibition of the hunger contractions. This fact was briefly reported in our first communication.[1] But Mr. V. is not an habitual smoker. It is therefore possible that the results obtained on him were simply due to the condition of nausea or disgust that smoking usually produces in the novice and hence not applicable to persons used to smoking.

The tests have now been repeated on several habitual smokers. *In so far as smoking influences the gastric hunger contractions this*

FIGURE 1. Tracing from the empty stomach of A. J. C. Bromoform manometer. Beginning of a period of hunger contractions, *x*, starting to smoke a "strong" cigar. Showing inhibition of the gastric hunger contractions and tonus.

influence is in the direction of inhibition. This inhibition appears to depend on the intensity of stimulation of the nerve endings in the mouth, a cigarette or "mild" cigar causing only slight inhibition, while a "strong" cigar or pipe causes complete and prolonged inhibition even when the gastric hunger contractions are at their maximum. A typical tracing showing this inhibition from smoking is reproduced in Figure 1.

If the cigar or pipe causes very strong stimulation of the nerve endings in the mouth, the inhibition of the hunger contractions may continue from five to fifteen minutes after the cessation of the stimulation. Thus even a brief period of smoking may suppress an entire hunger period.

[1] CARLSON: This journal, 1913, xxxi, p. 151.

The subjective sensation of hunger is diminished or abolished parallel with the gastric hunger contractions. But it seems to the authors that even a "mild" smoke diminishes the sensation of hunger rather more than one might infer from the slight depression of the contractions. This is probably due to the deviation of attention, the smoking acting partly as a "counter irritant."

Smoking inhibits the gastric hunger contractions. It is practically certain even in the absence of direct experiments that moderate smoking does not inhibit the gastric movements of digestion. The reason for the difference in the action of the same condition on the empty and on the filled stomach is not clear from present data.

II. The Influence of Constriction of the Belt

The experiments with constriction of the belt were made on three normal men. Mr. V. was not used in these tests for the

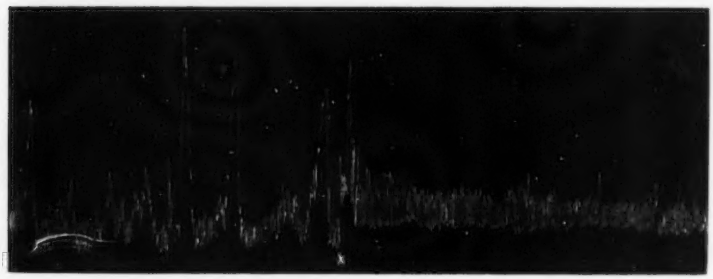

Figure 2. Tracing from the empty stomach of A. J. C. Beginning of a period of hunger contractions, *x*, strong pressure on the abdomen by belt. Showing inhibition of gastric hunger contractions of moderate strength by strong pressure on the abdomen by the belt.

reason that any considerable compression of the abdomen leads to pain and discomfort from pressure of the rubber tube in the gastric fistula. The tests were made with the subject standing up, sitting, and lying on the back, and at all stages of the gastric hunger contractions.

1. Strong contraction of the abdominal belt leads nearly always to inhibition of the gastric hunger contractions of weak or moderate strength, lasting from five to fifteen minutes. The inhibition may

be partial or complete, but in either case the hunger contractions reappear despite the continued pressure of the belt. This inhibition is obtained even when the belt constriction is moderate so that no discomfort or pain is produced. A typical tracing showing complete inhibition of the feeble hunger contractions on constriction of the belt is reproduced in Figure 2.

2. When the gastric hunger contractions are strong (the middle of a hunger period), constriction of the belt never causes complete inhibition. But so far as the increased abdominal pressure affects the hunger contractions the influence is in the direction of inhibition. The individual hunger contractions are weakened without

FIGURE 3. Tracing of the empty stomach of A. J. C. towards the end of a period of strong hunger contractions; *x-x*, strong pressure on the abdomen by belt. Showing a slight inhibition of the gastric hunger contractions during the compression of the abdomen. The hunger pangs were less intense during the time the belt was constricted.

suffering much change in the rate. Frequently, however, even a belt constriction that caused considerable discomfort has practically no influence on the hunger contractions, particularly if the subject is lying down. A tracing showing this slight inhibition is reproduced in Figure 3.

3. When the gastric hunger contractions are at their maximum in rate and amplitude, as is ordinarily the case near the end of a hunger period, no amount of belt constriction seems to influence the contractions. When this stage of the hunger period is reached the hunger pangs run their normal course in the presence of even painful belt pressures. A record illustrating this condition is reproduced in Figure 4.

4. All three subjects agreed that the belt constriction appeared to diminish or interfere with the hunger sensation to a greater

extent than seemed warranted from its effect on the hunger contractions. Several factors are probably involved in this discrepancy. (1) The belt constriction distracts the attention from the hunger impulses by stimulation of cutaneous nerves as well as by stimulation of nerve endings in the viscera, especially those of the peritoneum. (2) Strong pressure on the abdomen from without

FIGURE 4. Tracing from the empty stomach of J. H. L. (standing position). *X*, strong pressure on the abdomen by belt. Showing completion of the hunger period despite the belt constriction. But the period does not culminate in the incomplete tetanus characteristic for Mr. L.

appears to induce, temporarily, a condition simulating in a feeble way the complex sensation of satiety.[1]

[1] According to R. Lennhoff (quoted in Jour. Amer. Med. Assoc., 1913, lx, p. 41) hunger and appetite are appeased with a less quantity of food when the belt is constricted than when the intra-abdominal pressure is regulated solely by the tonus of the abdominal muscles. Lennhoff ascribes this to depression of hunger and appetite by the pressure of the belt. Lennhoff's observation is probably correct, but his explanation erroneous. The actual hunger contractions and the hunger sensations are stopped by the first few morsels of food swallowed, while this may actually increase the appetite through stimulation of nerve endings in the mouth and in the mucus membrane of the oesophagus and stomach. This appetite sensation is gradually counteracted by the sensation complex of satiety, which depends in part on the distention of the stomach with corresponding readjustment of the tonus of the abdominal muscles. This feeling of fullness, which appears to be referred to the abdomen as a whole, is probably developed with less intake of food when the abdominal wall is mechanically prevented from relaxing owing to the pressure of the belt.

5. We have practically nothing but conjectures to offer in way of explanation of the mechanisms involved in the above inhibition of the gastric hunger contractions by strong pressure on the abdomen. Strong pressure on the abdomen causes temporary inhibition of the gastric hunger contractions in dogs, but the manipulation greatly disturbs the dogs, and disturbance from any cause leads to a temporary inhibition of the empty stomach in dogs with the splanchnic nerves intact. In dogs with the splanchnic nerves sectioned on both sides, strong pressure on the abdomen causes no distinct inhibition of the gastric hunger contractions. This points to the conclusion that belt constriction causes gastric inhibition, not by direct pressure on the stomach, but by direct stimulation of inhibitory nerves, or by mechanical (or sympathetic) stimulation of the adrenal glands, but through long reflexes. Belt constriction involves stimulation of cutaneous nerve endings, but the stimulation of the tactile nerve endings in the skin alone does not lead to this inhibition. The afferent path of the reflex must therefore involve abdominal proprioceptors. The splanchnic nerves probably constitute the efferent path of the reflex. We do not wish to be understood as denying the existence of local inhibitory mechanism that may be stimulated by mechanical manipulation of the abdominal organs. But our results indicate that strong belt constriction is not a sufficient stimulus for such local mechanisms. In any event, belt constriction is not a very efficient control of the hunger mechanism. A "strong" cigar is more efficient in that direction than a good belt.

CONTRIBUTIONS TO THE PHYSIOLOGY OF THE STOMACH

XV. The Nervous Control of the Gastric Hunger Mechanism (Man, Dog)

By A. J. CARLSON

Assisted in the Experiments by J. H. Lewis and S. J. Orr

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THE activity of the gastric hunger mechanism is subject to reflex (nervous), and to chemical or "hormone," control. The present paper deals with the nervous control. The results of our studies of the control of the hunger mechanism through substances in the blood will be reported later.

Some data on the side of nervous control of the gastric hunger mechanism have already been reported. It has been shown that the nervous mechanism involves both local centres (Auerbach's plexus in the stomach wall) and the central nervous system. Stimulation of the sensory nerves in the mouth, oesophagus and stomach mucosa inhibits the hunger mechanism by way of the splanchnic nerves as well as through the Auerbach plexus. It has also been shown that practically all stimuli that act on the gastric hunger mechanism via the central nervous system cause inhibition, mainly through the splanchnic nerves. This is true, for example, in the case of the sight or smell of food on the part of dogs in hunger.

We now ask the reader's attention to the question of reflex control of vagus tone so far as this affects the stomach. We have determined the influence of the factors or conditions that are associated with lowering of the tonus of the central nervous system and the skeletal muscles, such as sleep, stimulation of the cutaneous nerve endings for heat, excessive muscular activity, as well as the factors that increase the skeletal neuromuscular tonus such as

stimulation of the cold nerve endings of the skin, moderate muscular activity, seeing and smelling palatable food, etc. These factors and conditions have been tested both on man and on dogs. Some of these conditions probably involve both chemical and nervous factors. Muscular activity may augment the gastric hunger activity by increasing the vagus tonus as well as by chemical changes in the blood. The same may apply to stimulation of the cold nerve endings of the skin. However, it is probable that if these conditions cause increase in the vagus tonus reflexly this response is more prompt than that induced by the changes in the blood following the increase or decrease in body metabolism due to stimulation. It is generally recognized that exercise, cold climate, and cold baths increase appetite and hunger. It does not follow that these conditions actually augment the gastric hunger contractions. The increase in hunger and appetite may be only apparent, that is, a condition of increased excitability of parts of the central nervous system, so that the afferent impulses that give rise to the sensations of hunger and appetite produce a greater central effect. If the gastric hunger contractions are actually increased, this may be due to changes in the blood rather than to increased vagus tonus.

It is well known that exposure of the skin to cold (as by bathing in ice water) may induce contracture or "cramps" of the digestive tract. This is especially the case during the height of gastric digestion. These cramps or contractures may be the result of circulatory disturbances or of changes in the blood rather than a direct reflex effect. Central processes are also able to induce contraction of the large intestine and the rectum, as shown by involuntary defecation in cases of great anxiety or fear.

From the point of view of biological adaptation we might expect the vago-gastric tonus to be directly affected by voluntary muscular activity and by exposure to cold, since both conditions involve increased oxidation and consequently increased need of food.

Experimental Procedure

Dogs. — Dogs with simple gastric fistulas were trained to run in a treadmill. When trained to run without much urging or

interference, records were taken of the contractions of the empty stomach so as to determine (1) whether muscular activity induces hunger contractions in the quiescent stomach, and (2) whether muscular activity augments the hunger contractions of an active stomach.

The hunger contractions of the stomachs of dogs were recorded for 2 to 4 hours, after a day's fast, the dogs being taken direct from the kennel without being exercised. On other days the same dogs were taken out for a 4 to 6 mile brisk walk before the 2 to 4 hour recording period.

Records of the gastric hunger contractions were taken with the dog lying quietly in the lap of an assistant. Then the body of the dog was surrounded with an ice pack, or the dog placed directly

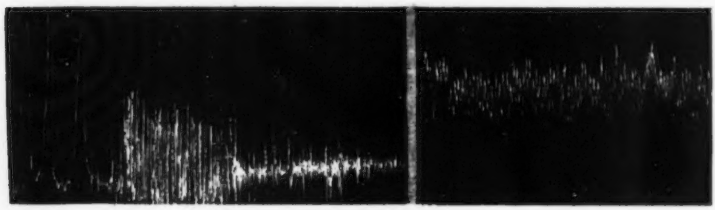


FIGURE 1. Tracing from the empty stomach of the dog. Bromoform manometer. *A*, dog standing in treadmill, stomach showing Type I hunger contractions. At *x*, the dog begins to run in the mill with the result that gastric hunger contractions are promptly inhibited. The running was kept up for 60 minutes. *B*, record from stomach of the same dog 45 minutes after he ceased running, showing increased gastric tonus and Type III hunger contractions.

on a slab of ice. After some training the dogs do not appear much disturbed by the ice pack or the slab of ice. The ice pack was applied with the stomach quiescent as well as in hunger activity.

All of the above procedures were used on normal dogs and on dogs with the splanchnic nerves sectioned on both sides, in order to have the tonus fibres of the vagi unopposed by the splanchnic inhibitory influence.

Man. — The tests were made on the author, on Mr. V. (the gastric fistula case), and on three assistants (J. H. L., S. J. O., A. M. P.).

Records were taken of the gastric tonus and hunger contractions with the man standing, and walking or running in place. Tests were also made after muscular exercise (playing tennis, walking 6 to 12 miles).

The influence of exposure to cold on the gastric hunger mechanism was tested in the following way. (1) While records of the gastric tonus and hunger contractions were being taken, the man, stripped of his clothes, was subjected to cold or warm showers for varying periods. The cold showers were at times sufficiently cold or prolonged to cause intense shivering. (2) The man stripped of his clothes in a cold room was covered up on a couch so as to feel comfortably warm. At the desired moment in the gastric activity, that is, during a period of quiescence or in the midst of a period of hunger contractions, the covers were removed and the cold air of the room set in motion by a fan placed close to the person. This brought on shivering in a few minutes. (3) The man arose at 7 A. M. and, without the usual cold bath and breakfast, proceeded to the laboratory and records of the gastric tonus and hunger contractions were taken from 8 to 12 A. M. These served as controls. On other days the man arose at 6 A. M., took a cold bath (this was prolonged until the discomfort became very severe), followed by a brisk walk, when records were taken from 8 to 12 A. M.

Results on Dogs

1. Effects of running in treadmill. — The initial effect on gastric tonus and hunger contractions of running in the mill is always in the direction of inhibition — usually complete inhibition, and if the dog is started running in the midst of a period of gastric quiescence there is no evidence of increased gastric tonus or beginning hunger contractions. If the dog is made to run at high speed the inhibition persists during the entire period even if the running is kept up for one to two hours. When the dogs ran at rather high speed for an hour or more the gastric inhibition usually persisted from 20 to 40 minutes after the dogs stopped running. The return of gastric tonus and hunger contractions in such cases is very gradual. But frequently when the gastric tonus finally recovered after a running period it was higher than before the dogs began to run.

Thus a dog showing Type I or II hunger contractions when he started to run in the mill may show an increased tonus and Type III hunger contractions 30 minutes after he stopped running, while the running period itself was accompanied by complete gastric inhibition. If the dog runs only moderately fast in the mill the gastric tonus and hunger contractions reappear during the running period, or come on during the running, in case the dog is started when the empty stomach is quiescent. A typical tracing showing this gastric hunger inhibition synchronous with running with subsequent recovery to greater gastric tonus is reproduced in Figure 1.

These facts indicate that the carnivorous animal in pursuit of its prey must be urged on by something else than the pangs of hunger, as these are inhibited by the chase.

2. Effects of 4–6 mile walk. — Eight tests (with a corresponding number of controls) on two dogs failed to show any marked effect of a 4 to 6 mile walk on the gastric hunger contractions either in the way of increase or decrease, the records being taken during the two hours following the walk. These walks certainly caused no depression of the dog's hunger contractions. But the dog that showed Type II contractions in the control usually showed Type II contractions after the walk with no definite increase either in rate or intensity. This should be noted, however, that after these walks both dogs showed greater restlessness than when taken from the kennels directly to the laboratory. They were not so easily quieted in the lap of the assistant. This rather restless condition of the dogs may have counteracted any augmentation of gastric hunger contraction due to the walk, as restlessness from any cause tends in the dog to inhibit the hunger contractions.

3. The effect of intense stimulation of the cutaneous nerve endings for the sensation of cold. — When a dog is lying quietly and comfortably in the lap of an assistant, surrounding the dog with an ice pack or placing him directly on a slab of ice leads to struggling and restlessness. After a number of repetitions of these procedures most dogs become so accustomed to it that they pay little or no attention to the change and show no restlessness or struggling. If the dog is disturbed or struggles when placed on a slab of ice or surrounded by an ice pack there always follows a temporary inhibition of gastric tonus and hunger contractions.

But this does not indicate the initial or primary effect of stimulation of the cutaneous nerve endings for cold, because the same type of inhibition is induced by restlessness or struggle for any cause. After the dog is trained to these procedures strong stimulation of the cutaneous nerve endings for cold by the ice pack, by placing the dog on a slab of ice, or by turning on an electric fan in a cold room after uncovering the dog, has no immediate effect on the gastric tonus and hunger contractions. There is usually an

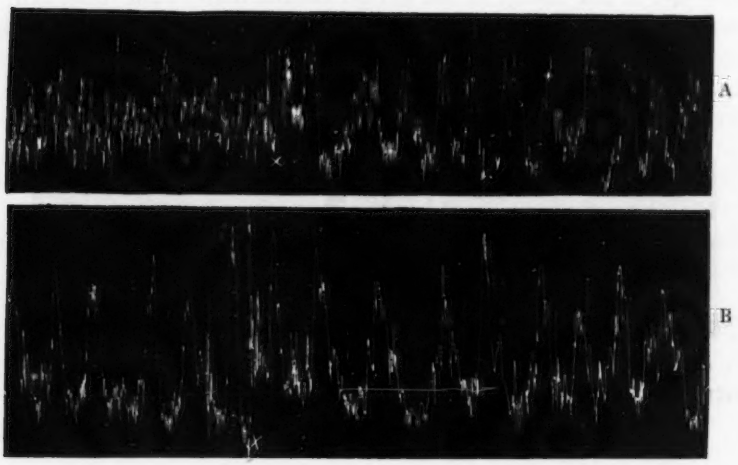


FIGURE 2. Tracings from the empty stomach of dogs. Bromoform manometer. *A*, dog covered with an ice pack for 30 minutes and shivering. Stomach shows Type III hunger contractions. At *x*, the ice pack is removed, and the stomach promptly passes into Type II hunger contractions. *B*, at *x* the dog was placed on a slab of ice. Showing no immediate effect on the gastric hunger contractions, despite the shivering of the dog.

increase in the intra-abdominal pressure owing to the increased tonus of the abdominal muscles. If the ice pack is applied during a period of gastric quiescence there is no immediate increase in gastric tonus or initiation of the hunger contractions, even though the dog starts to shiver violently in a few minutes. If the ice pack is applied during the hunger contractions, these contractions do not change appreciably either in rate or strength, at least for some time. This is true even when the dog shivers considerably. It would thus seem that the vagus centres governing the gastric

tonus are not directly affected by even very strong stimulation of the cutaneous nerve endings for cold.

In several instances the continued application of the ice pack (30 to 40 minutes) and in consequence continued shivering lead to a gradually increased gastric tonus and the appearance of Type III hunger contractions. These may be due to changes in the blood as a result of increased oxidation, or they may appear from causes not connected with the stimulation of the cold nerve endings. Such change in the hunger contractions is not infrequent in dogs, even when they are lying undisturbed and comfortable in the lap of an assistant.

In two cases the Type III hunger contractions changed to Type II on removal of the ice pack (Figure 2A). The change

x

FIGURE 3. Tracing from the empty stomach of dog with section of splanchnic nerves on both sides. At *x* the dog is surrounded by an ice pack. Showing no effect on the hunger contraction although the ice pack caused the dog to shiver.

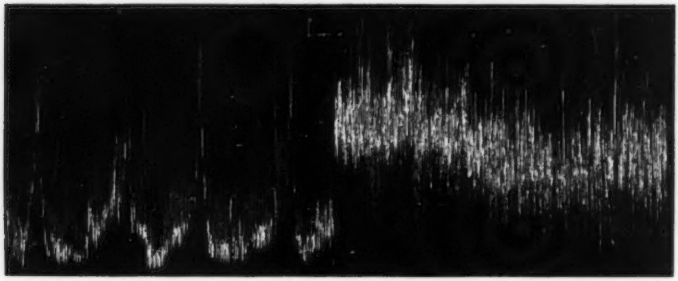
came on promptly on removing the ice pack. I am inclined to attribute this change to some shifting of the position of the stomach or shifting of the position of the balloon in the stomach as the result of the removal of the pressure of the ice pack over the abdomen, rather than as a reflex effect.

It is conceivable that the stimulation of the cold nerve endings in the skin does influence the vago-gastric tonus centres, but the stimulation acts equally on the gastric inhibitory mechanism via the splanchnic nerves so that the net result on the empty stomach is nil. This possibility is cleared up by the tests on dogs with section of both splanchnic nerves. Tests were made on two dogs on which this operation had been performed. The results were practically identical with those on normal dogs. The ice pack neither decreased nor increased the gastric hunger contractions

(Figure 3). It is therefore clear that the nervous impulses that give rise to the sensation of cold and induce increased neuro-muscular tonus in general have no direct action on the vago-gastric tonus centres.

Results on Man

1. The direct effect of muscular exercise. — Standing or walking in place has no effect on the gastric tonus or hunger contractions. But running in place promptly inhibits the hunger contractions (Figure 4). The degree and duration of the inhibition is on the whole directly proportional to the speed of the running.



x

Figure 4. Tracing from the empty stomach of man (A. J. C.) in standing position. Beginning of a hunger period. At *x* the man began running, with the result that the hunger contractions were promptly inhibited.

In some cases walking seemed to prolong a hunger period without changing the rate or intensity of the individual contractions. In no case did walking or running induce hunger contractions in the quiescent stomach. The results on man are thus identical with the results on dogs. In both species rapid running is accompanied by inhibition of the gastric tonus and hunger contractions. In the case of the dog running in the treadmill, one cannot be sure that the exercise is strictly voluntary and enjoyable. The inhibition may therefore be due to certain emotional states (anxiety, discomfort, mild anger or fear). This possibility is eliminated by the tests on man. In the men the conditions of the emotions when running in place were not different from that when standing or

walking in place. In no case was the running carried to the point of respiratory, cardiac, or muscular distress.

2. The after effects of muscular exercise. — Moderate exercise in the form of playing tennis or walking four to eight miles was taken in the afternoon. No supper was taken, and the motor condition of the empty stomach was recorded from 8 to 12 P. M. The tracings obtained on the days specific exercise was taken show on the whole greater gastric hunger activity than the controls. The periods of quiescence become shorter. This tends to make the gastric hunger contractions more or less continuous, and there appears to be some increase in the rate of the contractions. A typical experiment (S. J. O.) may be cited in the way of illustration.

Record of control day. — Lunch 1: 30 P.M. No special exercise. No supper. Period of observation 8 to 12 P.M.

8 to 10 P.M. Stomach practically quiescent.

10 to 10: 40. Strong hunger contractions ending in tetanus.

10: 40 to 11: 35. Stomach quiescent.

11: 35 to 12: 05. Moderate hunger contractions ending in tetanus.

Record of exercise day. — Lunch 1: 30 P.M. No supper, tennis 4 to 5 P.M.; walking 6 to 7 P.M. Period of observation 8 to 12 P.M.

8: 15 to 9: 50. Practically continuous hunger contraction ending in strong tetanus.

9: 50 to 10: 20. Stomach quiescent.

10: 20 to 11: 40. Strong hunger contractions ending in tetanus.

Total duration of hunger periods from 8 to 12 P.M. — Control day; 70 minutes. Exercise day; 190 minutes.

In some instances there was no marked difference between records of the control and the exercise days. This is to be expected since the activity of the gastric hunger mechanism depends in part on factors not understood or controlled. Exercise that brings on a degree of fatigue bordering on exhaustion seems to depress the gastric hunger mechanism. But our experiments on this point are as yet too few to permit a final conclusion.

3. The direct effect of stimulation of the cold nerve endings of the skin. — The immediate effect of stimulation of the cold nerve endings of the skin by ice pack, alcohol bath, cold shower bath, or

cooled air is inhibition of the gastric tonus and hunger contractions (Figure 5), and the degree of inhibition is proportional to the intensity of the stimulation. In no instance did we observe an initial increase in gastric tonus and hunger contractions. When the stimulation is continued the inhibitory effects gradually diminish even though the man shivers intensely from the cold. In this way the gastric hunger contractions may return to their normal rate, intensity and regularity, while the man is shivering and jerk-

X X′

FIGURE 5. Tracing from the empty stomach of man (A. J. C.) in the midst of a period of hunger contractions. The man was stripped and covered up with blankets in a cold room (20° C.). At *x* the covers were removed and a fan close to the man started. Shivering began at x^1. Showing a temporary but partial inhibition of the hunger contractions.

ing like a dog in mild parathyroid tetany. It may be noted in this connection that mild, and in some instances fairly severe, parathyroid tetany in dogs does not appreciably influence the gastric hunger contractions.[1]

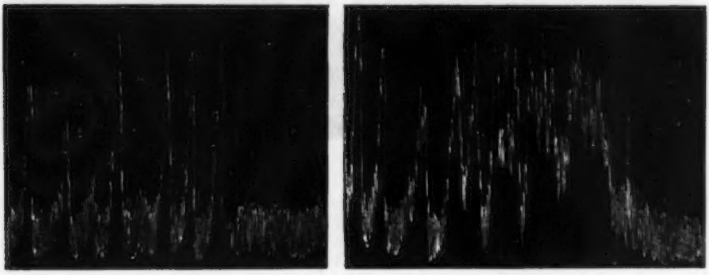
Intense stimulation of the heat nerve endings of the skin (hot shower) produces practically the same initial inhibition as the corresponding stimulation of the cold nerve endings.

While it is true that on prolonged stimulation of the cold nerve endings of the skin during a period of gastric hunger contractions, the inhibitory effects gradually disappear so that the contractions reappear in their normal intensity, these contractions are always felt as weaker than the normal, or may not be felt at all. Evidently the intense sensation of cold dominates consciousness to the exclusion of the gastric hunger pangs.

It is well known that strong stimulation of the cold nerve

[1] CARLSON: This journal, 1913, xxxii, p. 397.

endings of the skin causes a reflex increase of tonus of the urinary bladder. In several instances we started these stomach tests on the men at a time when their bladder was known to contain 50 to 200 cc. of urine. This permitted us to compare the reflex effect of cold on the stomach and on the bladder tonus without a balloon in the bladder. When the cold stimulation began during a period of gastric quiescence and was continued long enough to induce intense shivering a strong desire to micturate soon developed while there was no evidence of increased gastric tonus. Prolonged cold stimulation may produce so great tonus of the bladder that micturition cannot voluntarily be inhibited. The tonus centres



A B

FIGURE 6. Records of culminations of periods of gastric hunger contractions of A. J. C. *A*, the ordinary type of ending of the hunger periods without tetanus. *B*, hunger period ending in incomplete tetanus three hours after intense stimulation of the cold nerve endings (bath at 10° C. for 15 minutes).

of the urinary bladder are, the vago-gastric tonus centres are not, directly influenced by cold stimulation of the skin.

When the cold nerve endings of the skin are stimulated, as above, during a period of quiescence of the empty stomach, the stomach remains quiescent. If there is any change in the gastric tonus it is in the direction of inhibition. Nevertheless, this cold stimulation, if not sufficiently intense to be painful, seemed to induce a "sensation of emptiness" in the abdominal region, a sensation that seemed to be associated with appetite and desire for food. We record this with some hesitation, for this sensation of emptiness may be purely subjective (autosuggestion). It may also be due to the increased tonus of the abdominal muscles. In any event, this sensation is clearly different from the hunger pangs.

4. The after effect of stimulation of the cold nerve endings of the skin. — All of the tests in this group were made on one man (A. J. C.). A prolonged cold bath 6 to 7 A. M. followed by a brisk walk nearly always resulted in increased hunger activity of the stomach as recorded for the period 8 to 12 A. M. (Figure 6). The temperature of the water varied from 5° C to 15° C. The subject remained in the water as long as was deemed safe (10 to 20 minutes), despite discomfort and pain. Water at this temperature soon brings on shivering, contracture and at times severe headache, and it requires much vigorous exercise to restore the feeling of warmth. Rubbing the skin (rough towel) seems to be of no aid.

A typical experiment may be cited in illustration.

Control record. — No bath or breakfast. Observation period 8 to 12 A.M.

8: 50 to 10. 26 fairly strong hunger contractions; no tetanus.

11: 00 to 11: 45. 22 fairly strong hunger contractions; no tetanus. Gastric tonus on the average 5 cm. Bromoform.

Test period. — 6 to 6: 15 A.M. cold bath (temp. of water 10° C.). No breakfast. Observation period 8 to 12 A.M.

8 to 9. 32 strong contractions; no tetanus.

9: 45 to 10: 25. 23 fairly strong contractions; no tetanus.

11: 15 to 11: 45. 19 strong contractions ending in tetanus. Gastric tonus on the average 8 cm. Bromoform.

Control period. — 48 hunger contractions; no tetanus.

Test period. — 74 hunger contractions; tetanus.

Under ordinary conditions the periods of gastric hunger contractions of the author do not end in tetanus, but the hunger tetanus appears after 3 to 4 days' complete starvation.[1] Fifteen to thirty minutes' intense stimulation of the cold nerve endings thus seem to bring about a condition similar to prolonged starvation. This is in harmony with the observation of Lusk that such stimulation quickly renders the liver free from glycogen.[2] This effect of cold on the gastric hunger mechanism is obviously an indirect one, or through changes in the blood, and not a direct reflex from the skin.

[1] CARLSON: This journal, 1914, xxxiii, p. 95.

[2] LUSK: This journal, 1911, xxviii, p. 427.

Lusk has shown that intense cold leads to quicker and more complete oxidation of the body glycogen than prolonged starvation. And it is interesting to note that the same stimulus causes not only an increase in the *gastric hunger contractions*, but also an even greater increase in the subjective hunger and appetite sensations, probably owing to an increased excitability of the central nervous system. The increased desire to eat after a cold bath, in the case of the healthy individual, is a universal experience. I have investigated this matter in the case of young children, with whom habit or intelligence cannot be assigned as the cause for seeking food after a cold bath. It was found that young children react in the same way as adults.

5. The gastric hunger contractions during sleep. — During sleep there is decreased activity of the central nervous system in general, decreased tonus of the skeletal muscles, decreased tonus of the musculature of the blood vessels, at least in certain parts of the vascular system, decreased tonus of the urinary bladder, etc.; in short, a lowered activity of all the neuro-muscular mechanisms so far investigated. One might have expected that in so far as the tonus of the empty stomach depends on a central influence by way of the vagi, the gastric tonus and hunger contractions should be diminished during sleep. But instead of being depressed in sleep the hunger contractions continue with the same vigor as during the waking state, and in many instances with increased vigor. This has been established both for man and dog and is reported in previous communications.[1] We have nothing new to add on this point, except the mere confirmation of the facts already published. It is referred to in this connection because of its bearing on the question before us, the control of the vago-gastric tonus mechanism. The increase in the gastric hunger contractions during sleep may be due to elimination of all inhibitory impulses via the splanchnic nerves. But the absence of depression certainly indicates that the vago-gastric tonus mechanism, at least in man and dog, occupies a unique position in the organism, a degree of independence of afferent impulses (exteroceptors) and central processes not known in the case of any other neuro-muscular apparatus.

[1] CARLSON: This journal, 1913, xxxii, p. 369; 1914, xxxiii, p. 95.

6. The effect of cerebral states (emotional states, Intellectual processes). — It has been shown in previous communications that in the dog the nervous processes of joy, fear, anger, eagerness (from food), attention, etc., cause temporary inhibition of the gastric hunger contractions. This inhibition takes place by way of the splanchnic nerves, not by a depression of the vagus tonus. This, again, points to an unusual independence of the vago-gastric tonus apparatus.

In man, intellectual processes (attention, reading, figuring, arguing) have no distinct influence on the course of the hunger periods. Actual anxiety causes temporary inhibition (probably through the splanchnics). We have not been in position to make observations on the effects of actual anger, fear and joy, but there is no reason to believe that these processes act differently in man from that in the dog. In man we have paid particular attention to the effects of seeing and smelling palatable food, as it seemed *a priori* reasonable that the impulses generated by these stimuli might make more intimate connection with the vago-gastric tonus apparatus. Extensive experiments on Mr. V. and a number of tests on the author seem to show that this is not the case. These stimuli neither initiate nor augment the gastric tonus and hunger contractions; so far as they influence them at all, it is in the direction of inhibition. One of the tests on the author might be given. Before beginning the five days' starvation period, our colleague, Dr. Luckhardt, was asked to bring in, unknown to the author, a tray of choice food in the midst of a hunger period. The arrangements being made, the matter was dismissed from the author's thoughts.

One o'clock on the morning of the fourth starvation day the subject was asleep and the record showed the midst of a period of vigorous and regular hunger contractions. He was awakened to behold Dr. Luckhardt and the assistant enjoying a feast of porter-house steak with onions, German fried potatoes, and a tomato salad. The tray with edibles was placed not more than four inches from the subject's face and the delicious odor of the food filled his nostrils. He felt the hunger pangs as unusually intense, and there was considerable salivation. However, the gastric hunger contractions were not increased either in rate or intensity.

In a few minutes, on the contrary, the hunger contractions became weaker and the intervals between them greater, and the period terminated by this gradual depression much sooner than it probably would have done in the absence of the dinner scene. This was undoubtedly due to local acid inhibition from copious secretion of appetite gastric juice.

Our data on normal men and dogs seem incapable of any other interpretation than that the vago-gastric tonus apparatus so far as it concerns the empty stomach occupies a unique and physiologically isolated position, in the way of nervous control, while the inhibitory apparatus via the splanchnic nerves is readily influenced by central and reflex processes. We feel, however, that these observations must be extended to other groups of vertebrates as well as to such pathological cases in man in which there are indications of abnormalities of the vago-gastric tonus, before final explanations are attempted or speculation indulged in as to the usefulness of this physiological isolation.

This evidence for the physiological isolation of the hunger mechanism in the way of positive central control is of interest in connection with the view that the cravings of hunger and appetite are subjective and largely a matter of habit, and that the periodicity or intensity of these cravings may be altered almost at the will of the individual. Chittenden[1] states this view as follows. "The so-called cravings of appetite are largely artificial and mainly the result of habit. Anyone with a little persistence can change his or her habits of life, change the whole order of cravings, thereby indicating that the latter are essentially artificial and have no necessary connection with the welfare or needs of the body. The man who for some reason deems it advisable to adopt two meals a day in place of three or four, at first experiences a certain amount of discomfort, but eventually the new habit becomes a part of the daily routine, and the man's life moves forward as before, with perfect comfort and without a suggestion of craving, or a pang of hunger."

Our studies of the hunger mechanism seem to show that the above view is essentially wrong. In the normal individual the gastric hunger periods begin as soon as the stomach is empty and

[1] CHITTENDEN: The Nutrition of Man, New York, 1907, p. 164.

continue (in the absence of inhibitory processes) as long as the stomach is empty, irrespective of the time of day or night, and without reference to the time the individual is accustomed to eat. In individuals accustomed to the usual three meals in daytime and to sleep during the night, the gastric hunger periods are more frequent and usually more vigorous during the night (in sleep) than during the day, provided, of course, the stomach is empty. In the normal individual the empty stomach exhibits periodic hunger activity, and there is no evidence to show that this primary automotism of the empty stomach is in the least influenced by eating one or by eating five meals a day. The basis for the view that the time of appearance of the "cravings of hunger" can be changed at will is probably to be sought in the fact that the milder hunger contractions do not enter consciousness as pangs of hunger if the individual's attention is directed into other channels. They are felt as hunger pangs if the individual's attention is directed towards food and eating. The attention is thus directed, consciously or sub-consciously, about the time the individual is accustomed to eat. The periodicity of this subjective attention to the milder hunger cravings can probably be altered by training. But this applies only to relatively mild pangs of hunger. The more severe "cravings of hunger" caused by the gastric hunger tetanus rise above the limen of consciousness, except in deep sleep or under conditions of cerebral process involving intense interest. When an individual who is used to eat three times a day turns to a régime of one meal a day, the quantity of food ingested in that one meal is much greater than that at any one of the three meals a day régime. The emptying of the stomach and the appearance of the pangs of hunger are correspondingly delayed. The view that prompt appearance and the persistence of the gastric hunger activity in the empty stomach have no relation to the actual need of the individual for food cannot be seriously maintained for the normal animal.

Summary

1. Moderate muscular activity (walking) has no direct influence on the gastric hunger mechanism. Intense muscular activity (running) inhibits the hunger mechanism in direct proportion

to the intensity and duration of the exercise. Feeble hunger contractions may continue during moderate running. There appears to be some increase in the gastric tonus and hunger contractions as an after effect of moderate exercise.

2. Stimulation of the cold nerve endings of the skin does not affect the vago-gastric tonus apparatus. If the stimulation is of sufficient intensity it induces (especially in man) a temporary inhibition of the gastric hunger contractions via the splanchnic nerves. A similar inhibition is induced by strong stimulation of the cutaneous nerve endings for warmth. There is a distinct increase in the gastric tonus and hunger contractions as an after effect of prolonged and intense stimulation of the cold nerve endings of the skin.

3. The vago-gastric tonus mechanism is not influenced by the condition of sleep, except in the way of augmentation, owing to the elimination of all inhibitory processes via the splanchnic nerves.

4. The vago-gastric tonus mechanism is not affected by intellectual processes or emotional states, except in so far as these cause inhibition of the gastric tonus and hunger contractions via the splanchnic nerves.

5. It is clear from the above that in normal individuals (man, dog) the vago-gastric tonus apparatus, at least so far as it concerns the empty stomach, is physiologically isolated from the exteroceptors and from many, if not all, central processes, while the splanchnic inhibitory apparatus is readily accessible to these processes. The biological significance of this exceptional and unique isolation of the tonus apparatus of the hunger mechanism probably lies in the importance of the hunger mechanism being regulated on its positive side primarily by the state of nutrition, that is, through the blood, rather than by the fleeting changes in the nervous system.

ADRENAL DEFICIENCY AND THE SYMPATHETIC NERVOUS SYSTEM

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ELLIOTT'S final demonstration of the intimate relationship subsisting between epinephrin and the sympathetic nervous system [1] has seemed to many physiologists largely to have solved the problem of the functional significance of the adrenal glands. The fact that injecting epinephrin is exactly equivalent to a general stimulation of the sympathetic system and the common supposition that an animal after removal of its adrenals dies in a condition of vasomotor failure at least strongly suggest that the paramount function of these glands is to maintain tonus in the sympathetic system. This hypothesis may be designated the "tonus theory." It supposes that the adrenals constantly pour into the blood stream epinephrin in sufficient amount to keep the sympathetic system in a condition of partial stimulation.

A number of facts may be mentioned, however, which seem to disprove the theory. Blood pressure which is directly under sympathetic control may be taken as a criterion of sympathetic functioning. Experiments have shown that epinephrin at a sufficiently slow rate can be introduced continuously into a vein without producing any demonstrable effect.[2] If the rate is increased to an effective degree the result supposedly is either an increment of the influence of the epinephrin normally derived from the animal's own glands or else, if that has been in abeyance, of the appearance of the normal physiologic epinephrin effect. But the first apparent

[1] ELLIOTT: Journal of Physiology, 1905, xxxii, p. 401.

[2] HOSKINS and MCCLURE: Archives of Internal Medicine, 1912, x, p. 343.

result of such injections is a *depression* of vascular tonus.[1] This is Elliott's "paradoxical reaction."[2] Whether, however, this be the paradox, is merely a matter of orientation; the pressor effect of larger doses might equally well be so designated. As a matter of fact a reversal of reaction as the acting quantity of epinephrin is increased seems characteristic. Such reversals have been observed in case of intestinal peristalsis,[3] uterine contractions,[4] pulmonary circulation[5] and general blood pressure[1]. It is the effects observed with the higher dilutions that are probably to be regarded as normal.

Another fact militating against the "tonus" theory is that sudden ligation of the adrenal circulation under conditions whereby nothing else is affected has absolutely no influence upon blood pressure until the lapse of a period far greater than that required for the destruction of any accumulated epinephrin that might have been present. The matter has been conclusively determined both in anaesthetized and in conscious animals.[6] If the theory were true a fall of pressure exactly commensurate with the preceding tonic influence would necessarily occur immediately after the ligatures were placed.

Possibly most significant is the fact that the injection of any quantity of epinephrin adequate to exert a minimal "tonic" influence upon blood pressure gives rise to conditions incompatible with ordinary existence. In dogs, such quantities produce complete paralysis of the gastrointestinal tract.[7] In rabbits also this

[1] MOORE and PURINGTON: Archiv für die gesamte Physiologie, 1900, lxxxi, p. 483. HOSKINS and MCCLURE: Archives of Internal Medicine, 1912, x, p. 353. CANNON and LYMAN: This journal, 1913, xxxi, p. 376.

[2] ELLIOTT: Journal of Physiology, 1912, lxiv, p. 402.

[3] HOSKINS: This journal, 1912, xxix, p. 363.

[4] STEWART: Journal of Experimental Medicine, 1912, xv, p. 547.

[5] DESBOUIS et LANGLOIS: Comptes rendus de la Société de Biologie, 1912, lxxii, p. 674.

[6] HOSKINS and MCCLURE: This journal, 1912, xxx, p. 192. KAHN: Archiv für die gesamte Physiologie, 1911, cxl, p. 216. TRENDELENBURG, W: Zeitschrift für Biologie, 1914, lxiii, p. 155.

[7] HOSKINS and MCCLURE: This journal, 1912, xxxi, p. 59.

is true of some individuals.[1] Moreover, during such injections a condition of glycosuria arises before a pressor effect appears.[2]

Attractive as the tonus theory is, such data render it no longer tenable. But the fact remains, — adrenal extirpation is fatal and the final symptoms are supposed to include a primary failure of functions under sympathetic control, — notably of blood pressure. Elliott[3] has offered the suggestion that adrenal deficiency results, not necessarily in the loss of any tonic stimulant, but of a substance necessary for the maintenance of sympathetic irritability; that is, that epinephrin is of importance in the metabolism of the sympathetic system or more particularly of the myoneural "receptive substance." There is nothing in the available evidence which disproves the suggestion.

Another hypothesis equally capable of explaining the results of adrenal extirpation has been made by Hoskins and McClure,[4] — namely that the adrenals in some way directly promote the metabolism of the muscular tissues. In the absence of the glands myasthenia develops. This asthenia, if it included the circulatory apparatus, would lead to low blood pressure. The negative phase of the hypothesis is equally tenable, that is, that the adrenals destroy some substance which interferes with muscular metabolism. In this form the suggestion is an old one.

Elliott's suggestion is amenable to experimental investigation. If adrenal destruction results in an interference with sympathetic functioning the fact should be easily demonstrable. Accordingly the experiments herein reported were undertaken.

At first thought, an animal in a late stage of fatal adrenal deficiency might seem to offer the most favorable conditions for determining the matter. But such can scarcely be the case. An animal in the final stages is simply moribund and can afford little definite evidence as to how it became so. The classic description of the effects of adrenal extirpation is not at all significant. It is nothing more than the description of an animal dying from any

[1] TRENDELENBURG, P., und FLEISCHHAUER, K: Zeitschrift für die gesamte experimentelle Medizin, 1913, I, p. 393.

[2] GRAMENITZKI: Biochemische Zeitschrift, 1912, xlvi, p. 186.

[3] ELLIOTT: Journal of Physiology, 1904, xxxi, p. xx.

[4] HOSKINS and MCCLURE: This journal, 1912, xxx, p. 195.

non-irritative slowly cumulative cause; muscular weakness, subnormal temperature, feeble respiration and pulse, low blood pressure — none of these are at all characteristic and any or all might be secondary effects. More significant are the conditions when the animal is first reacting to the operation. Primary effects alone are then in evidence. Our studies were made, therefore, mostly on earlier stages, from two to six hours after removal of the glands. In one instance, however, an interval of nine hours elapsed and one animal under urethane anaesthesia was kept under continuous observation from the time of operation until death, about ten hours later.

Various means of investigating the condition of the sympathetic system were contemplated but only three were found necessary. These are stimulation of afferent nerves, and injections of "adrenalin" and of nicotin. Blood pressure was used as a criterion of sympathetic conditions and by these three means conclusive evidence as to the conditions of the vasomotor reflex arc were secured before and after adrenal destruction. As a matter of fact the epinephrin, as the final results showed, might have been omitted, but this fact was not foreseen. We made observations in each case also with pituitrin but these added nothing significant to the evidence and are not reported.

In all the experiments dogs were used. The plan of procedure in most cases was to make a preliminary study of vasomotor conditions, remove the adrenals, and after the animal began to react to adrenal deficiency, make a second investigation of the vasomotor system. In some instances the removal of the adrenals immediately preceded the first determination. Under ether anaesthesia the animals were prepared for recording blood pressure. Into a femoral artery was inserted a Hall reservoir cannula[1] which was filled with 10% sodium citrate solution and connected directly with a recording mercury manometer. Into the contiguous vein was tied a large-bore cannula which was connected by rubber tube with a reservoir of 0.8 per cent sodium chloride solution suspended at a height of two feet. The effects of nicotin and of adrenalin vary materially according to the speed at which they are

[1] The Hall cannula is an ordinary glass arterial cannula in which is blown a 20 cc. reservoir.

introduced. To secure uniformity in this regard the following technique was employed: The substance was injected by a hypodermic syringe into the rubber tube just above the venous cannula. Its entrance into the cannula was prevented by a clip. Immediately then before diffusion occurred the clip was released and the drug instantaneously flushed into the vein. Previous investigation had shown that this technique gives results at different times which are comparable.[1]

In the leg not used for the cannulas a crural nerve was laid bare for stimulation. For this the nerve was simply picked up on platinum electrodes. In order to obviate local injury of the axones and the effects of variability of contact at different times, the electrodes during the time of stimulation were moved back and forth over a segment about two centimeters long. A strength of current was employed just sufficient to give a definite pressor response. The same strength of course was used in all experiments in any given animal. By these means blood pressure records were obtained showing the initial degree of vasomotor tonus and of vasomotor irritability. They gave an index also of the strength and rapidity of the heart beat. Some knowledge of respiratory conditions could be deduced from the respiratory waves of the pressure curves. At the conclusion of the determinations the vessels were tied off and the incisions in the legs closed with sutures. In the interim between experiments the animals were kept lightly under the influence of morphine. In the succeeding determinations the same blood vessels and nerves and the same cannulas were used as at first.

That complete ligation of the adrenal glands is equivalent to their actual extirpation has been shown by Mossou and Le Play [2] and confirmed by Allen.[3] Animals die with the same symptoms after either procedure. Ligation has the material advantage that it can be carried out in ten minutes and with a minimal degree of shock. We decided, therefore, to use this method of creating adrenal deficiency. In order to be certain of the absolute isolation

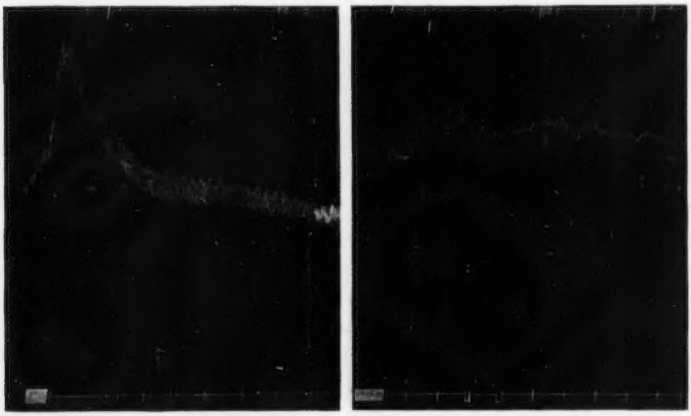
[1] Hoskins and Wheelon: This journal, 1914, xxxiv, p. 82.

[2] Mossou et Le Play: Comptes rendus de la Société de Biologie, 1909, p. 36, p. 83.

[3] Allen: Glycosuria and Diabetes: Boston, 1913, p. 863.

of the glands two ligatures were introduced together under each. One ligature was then tied tightly at the mesial, the other at the lateral side. All possible connection with the rest of the body was thereby destroyed. Our actual results show that diffusion from the isolated organs is not a factor. The animals promptly developed the characteristic signs of deficiency just as after actual extirpation.

Figure 1 shows the reactions of dog 51 to 0.4 cc. of 1:20,000 solution of "adrenalin" before and after adrenal ligation. The



A B

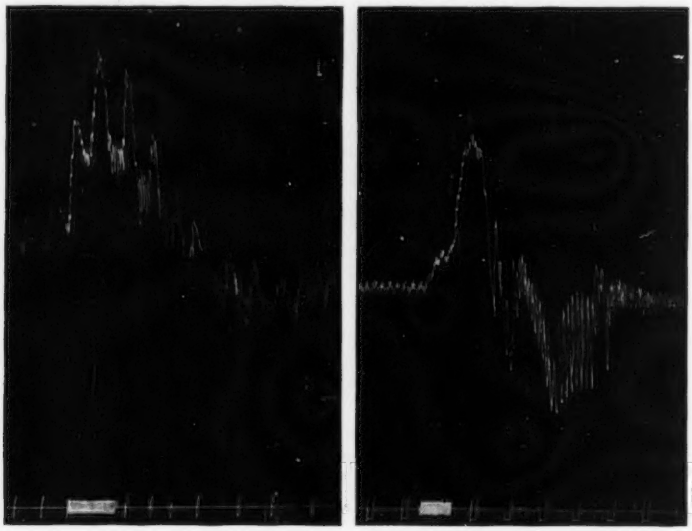
FIGURE 1. Dog 51. (*a*) 1:45 P.M. Reaction to 0.4 c.c. adrenalin, 1:20000. (*b*) 5:58 P.M. Reaction to same quantity adrenalin. Time, 5 sec. Adrenals ligated 2:10 P.M.

first record (*a*) was secured at 1:45 P.M. At 2:10 the adrenals were ligated. The animal recovered promptly from the anaesthetic but showed some evidence of shock. It remained more quiet than a dog which had been merely under either for the same length of time. Ultimately, however, it got up and walked about. At 5:58, about four hours after the first determinations, the reaction to 0.4 cc. of adrenalin was again taken. The blood pressure was found but slightly lower than in the initial case. The reaction to adrenalin was approximately the same. The most notable difference in the two records is in the amplitude of pulse. The heart

beat has become strikingly weaker than before the adrenals were removed.

Figure 2 shows somewhat similar conditions in another animal. The interval between the determinations was $4\frac{1}{2}$ hours. The reaction to adrenalin was slightly increased above normal. The record shows the decrease in respiratory waves that was characteristic of all the experiments.

Figure 3 shows three similar determinations but all made after adrenal ligation. The intervals are approximately $\frac{1}{2}$, $6\frac{1}{2}$ and 9 hours after the glands were tied off. The animal at the time of



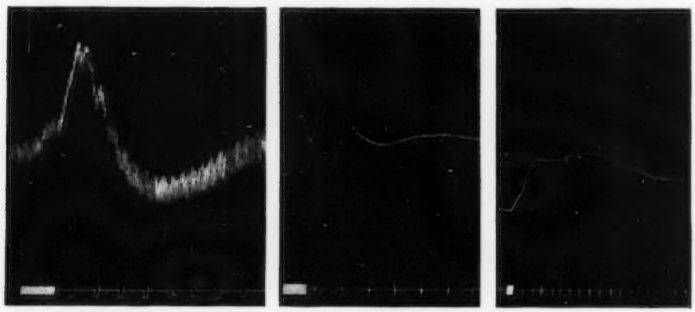
A B

FIGURE 2. Dog 47. (*a*) 11:00 A.M. Reaction to 0.4 c.c. adrenalin, 1:20000. (*b*) 4:30 P.M. Reaction to same quantity adrenalin. Time, 5 sec. Adrenals ligated 10:35. A.M.

the last determination was nearly moribund. It had shown extreme muscular weakness throughout the latter part of the experiment. The records show a persistence of vasomotor tonus and of irritability to adrenalin with the same marked weakening of the heart as in the preceding cases. It is difficult to understand how even with the greatest possible vasomotor efficiency the pressure

of the last determination could have been maintained with a heart too weak to cause an appreciable pulse wave. Figures 1, 2 and 3 show that there is little or no loss of vasomotor tonus or of irritability of the myoneural "receptive substance" even in an animal showing evidence of extreme adrenal deficiency.

Figure 4 shows the reaction of dog No. 50 to 0.8 cc. of 1:2000 nicotin shortly after adrenal ligation and again 4 hours after. It shows the same weakening of heart beat and respiration as in the preceding records and an *augmented irritability to nicotin*. If, as



A B C

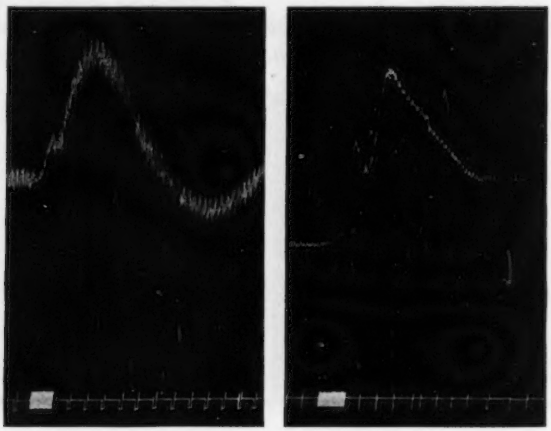
FIGURE 3. Dog 49. (*a*) 10:45 A.M. Reaction to 0.4 c.c. adrenalin, 1:20000. (*b*) 4:58 P.M. Reaction to same quantity adrenalin. (*c*) 7:05 P.M. Reaction to same quantity adrenalin. Time, 5 sec. Adrenals ligated 10:20 A.M.

Langley states,[1] the stimulating effect of nicotin is chiefly upon the sympathetic ganglion cells, the observation would indicate that adrenal extirpation results in an augmented irritability of the peripheral sympathetic system. A similar result was noted in other but not all cases. In one instance it was true to a much greater degree. Even though such augmented irritability were characteristic in all cases it might often be masked by the concomitant cardiac weakness.

Figure 5 shows the reaction to nicotin persisting in a nearly moribund animal, nine hours after ligation of the adrenals. Considering the cardiac weakness the extent of reaction is remarkable. It would indicate that there is certainly no loss of sympathetic irritability involved in the symptom complex of adrenal deficiency.

[1] LANGLEY and DICKASON: Journal of Physiology, 1890, xi, p. 297.

Figure 6 shows the reaction of dog No. 49 to sensory stimulation a half hour and nine hours after adrenal ligation. The stimulus was a Faradic current applied as previously described. The secondary coil was in the same position in each case, a distance of 8 cm. from the primary, — and the primary current was the same, — one dry cell. The observation indicates that the whole vaso-



A B

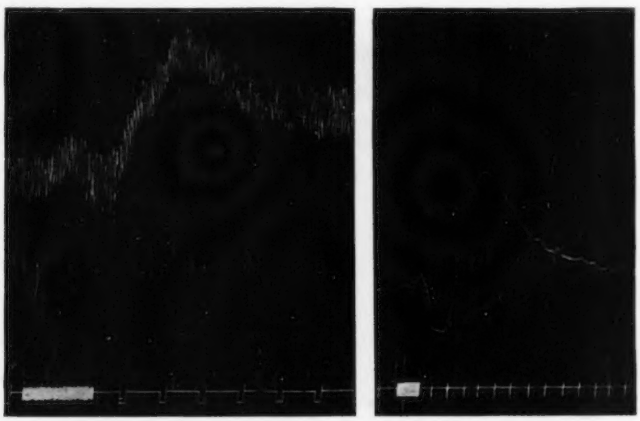
FIGURE 4. Dog 50. (*a*) 11:27 A.M. Reaction to 0.8 c.c. nicotin, 1:2000. (*b*) 3:36 P.M. Reaction to same quantity nicotin. Time, 5 sec. Adrenals ligated 11:05 A.M.

motor arc retains its function at a time when adrenal deficiency has reached an extreme degree.

The results of the series as a whole were consistent and convincing. At a time when the animal showed clearly the characteristic muscular weakness of adrenal deficiency amounting almost to complete paralysis of the hind limbs the vasomotor system was not in the least impaired. As the condition progressed the heart beats became remarkably weak, but a significant lowering of blood pressure did not appear until a late stage. This fact shows that some sort of compensatory reaction must have occurred. That this reaction is an augmented sympathetic irritability is specifically indicated by several nicotin experiments. The lowered blood pressure that finally developed is to be ascribed to primary cardiac failure.

The results obviously show also that the smooth muscle of the arterioles and capillaries does not share in the general myasthenia. This fact which was quite unexpected made the solution of our problem easier than we had hoped.

That shock is not a factor in our experiments is shown by the fact that the results — as in Figures 1 and 3 — were the same



A B

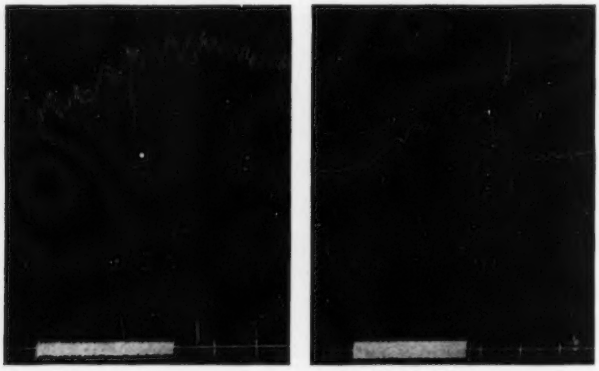
FIGURE 5. Dog 49. (*a*) 10:40 A.M. Reaction to 0.8 c.c. nicotin, 1:2000. (*b*) 7.03 P.M. Reaction to same quantity nicotine. Time, 5 sec. Adrenals ligated 10:20 A.M.

whether the initial determination preceded or followed ligation of the adrenals.

One fact was noted, the significance of which is not clear. In most instances the effects of the nicotin and the adrenalin injections persisted notably longer after ligation of the adrenals than before. Figures 1 and 3 show characteristic instances. Our data do not indicate whether this persistence is to be ascribed to a delayed destruction of the drugs, to a change in the reacting tissues or merely to a slowed circulation whereby the reacting tissues are longer exposed to the stimulating substance. The records as a whole indicate that the effect is not to be ascribed to improved heart action.

In view of the importance that has been ascribed to the adrenal as a "hypertensive" gland the literature shows a surprising dearth

of exact pertinent information. We have found but two accounts of preceding experimental investigations such as that herein reported. Elliott has stated in an abstract report that cats in a moribund condition after adrenal extirpation no longer react to nicotin. Gautrelet and Thomas[1] in 1909 reported in a short "comptes rendus" article the results of another investigation of sympathetic irritability after adrenal extirpation. They reached a conclusion



A B

FIGURE 6. Dog 49. (*a*) 10:50 A.M. Faradic stimulation Crural nerve. (*b*) 5:06 P.M. Stimulation same nerve with same strength current. Adrenals ligated 10:20 A.M. Time, 5 sec.

directly opposed to ours. Their results in brief are as follows: Faradic stimulation of the cervical sympathetic nerve on one side gave a mydriasis with the secondary coil of the inductorium at 10 cm. Five hours after adrenal extirpation a similar stimulation of the cervical trunk on the other side gave a mydriasis only when the secondary coil was at 7 cm. Similarly stimulation of afferent nerves or of the splanchnic trunk when the animal was under the influence of adrenal deficiency failed to affect blood pressure although the splanchnics had been proven irritable during the course of the operations upon the adrenals. They noted also in a rabbit a congestion of the ear which was influenced neither by heat nor

[1] GAUTRELET et THOMAS: Comptes rendus de la Société de Biologie 1909, p. 388.

by cold. They concluded, therefore, that within five hours after adrenal extirpation in both dogs and rabbits the sympathetic nervous system undergoes a loss of irritability. Their results obviously did not show whether the depression was in the nervous elements or in the effector mechanisms. The unfortunate brevity of their report renders a critical consideration of the work impossible. In the light of our experiments it can only be conjectured that their animals as well as Elliott's may have been so nearly moribund as to be incapable of giving any definite information. We have pointed out in a preceding paragraph the desirability of studying the problem in animals that have not yet reached such an extreme stage. It is to be noted that in dogs, as occurred in one of our own experiments, the animal may succumb within six hours of the extirpation.[1]

Schwartz[2] has made certain incidental observations that might be interpreted as bearing upon the problem under discussion. He has found that rats deprived of their adrenals although surviving in apparent good health, within two or three days acquire an augmented sensitiveness to epinephrin. The case of one animal is described particularly: Six weeks after epinephrectomy the animal was given subcutaneously 0.2 mg. of the drug, — a dose which in normal rats is ineffective. It developed extreme restlessness and dyspnea. It ran about the cage frothing at the mouth and with blood coming from the nostrils. Examination after death showed punctiform ecchymoses in all the serous membranes and extreme edema of the lungs, indicating vascular hypertension as the cause of death. The observation so far as it goes accords with our finding of increased sympathetic irritability.

Battelli,[3] in 1902, in a study of adrenal extirpation in which circulatory conditions were particularly under observation found sudden arrest of the heart to be the characteristic cause of death. This finding agrees with ours that cardiac weakness is a primary result of adrenal destruction.

At first thought our results might seem to accord with previous reports postulating a specific relationship between epinephrin and

[1] GRADINESCU: Archiv für die gesamte Physiologie, 1913, cli, p. 203.

[2] SCHWARTZ: *Ibid.*, 1910, cxxiv, p. 281.

[3] BATTELLI: Comptes rendus de la Société de Biologie, 1902, p. 1138.

striated muscle. Researches in Cannon's laboratory[1] have recently shown that a temporary improvement in the reactions of a fatigued muscle undoubtedly follows the injection or discharge of epinephrin. A simple explanation for our observations would be that adrenal extirpation simply reduces the quantity of circulating epinephrin below the minimal amount necessary to maintain muscular metabolism. Several facts, however, oppose such a theory: In the first place if *epinephrin deficiency* were the significant factor it could easily be compensated for by continuous intravenous infusion of the drug. But Battelli has shown that such procedure is absolutely futile. The survival time is either not at all affected or else is actually shortened. Gradinescu[2] in more recent experiments has found that occasional epinephrin injections prolong life somewhat but exert no more than a temporary benefit. The animals invariably die in any case. That there is such a thing as normally circulating epinephrin is pure assumption. That adrenal discharge occurs as a result of various unusual conditions there is no doubt,[3] but there exist no reliable determinations of an epinephrin content of blood collected under normal conditions. Epinephrin in detectable amount does not exist in arterial blood collected from a quiet animal by cardiac puncture.

Moreover, what direct evidence there is indicates that it is the loss of cortical, not chromaffin tissue that leads to the fatal issue of adrenal extirpation. Biedl's experiments upon selachians are well known.[4] Compensatory hypertrophy of adrenal fragments has often led to survival of an animal after extirpation of the glands, but in such cases it is the cortical tissue alone that hypertrophies. Weed[5] has recently reported a crucial experiment upon the point. In an animal which had been preserved by a surviving adrenal fragment ligatures were so placed that the circulation of the cortical part was destroyed, but the chromaffin moiety left unaltered. The animal promptly died. The greater liability to muscular

[1] CANNON and NICE: This journal, 1913, xxxii, p. 44. GRUBER, *Ibid.*, 1914, xxxiii, p. 335.

[2] GRADINESCU: loc. cit.

[3] CANNON: This journal, 1914, xxxiii, p. 356.

[4] BIEDL: Innere Sekretion. Berlin, 1913.

[5] WEED: Verbal communication before the American Society of Experimental Pathologists, Dec. 30, 1913.

fatigue after epinephrectomy observed by Albanese[1] and by Boinet[2] is equally explicable on either hypothesis. Dessy and Grandis'[3] report that application of epinephrin has a long-continued sustaining effect upon frog muscle one of us[4] has been unable to confirm. Moreover, the characteristic syndrome of Addison's disease may develop in patients in whom the cortical tissue alone is affected.[5] On the whole, therefore, it seems most likely that the brief beneficial effect of epinephrin upon striated muscle is but a part of its emergency function and that the characteristic myasthenia following adrenal extirpation is due to cortical deficiency.

Summary and Conclusions

1. Complete ligation of both adrenal glands of dogs at a single operation causes within 4 to 6 hours characteristic weakness of the skeletal muscles, — including those of respiration.

2. The weakness is shared to a marked degree by the cardiac muscle.

3. At a time when cardiac weakness is strongly in evidence blood pressure remains at or near its initial height.

4. A compensatory activity of the vasomotor system therefore occurs.

5. Vasomotor responses to Faradic stimulation of the crural nerve persist. The vasomotor reactions to adrenalin also persist undiminished. The reactions to nicotin are often somewhat exaggerated as compared with preliminary observations with the same dosages.

6. The vasomotor system therefore as well as the vascular musculature are unimpaired at a time when marked asthenia of skeletal and cardiac muscle has developed.

7. This asthenia is sufficient alone to account for the final fatal results of adrenal extirpation.

8. We find no evidence, therefore, that the sympathetic system suffers primarily in any degree from adrenal extirpation.

[1] Albanese: Archives italiennes de Biologie, 1892, xvii, p. 243.

[2] Boinet: Comptes rendus de la Société de Biologie, 1895, xlvii, pp. 273, 498.

[3] Dessy et Grandis: Archives italiennes de Biologie, 1904, xli, p. 223.

[4] Hoskins: Experiments not hitherto reported.

[5] Löwy: Deutsches Archiv für klinische Medizin, 1913, cx, p. 373.

THE RELATION OF PULSATION TO FILTRATION

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The Effect of Pulsation on Filtration

THE effects of vibration on living protoplasm and on colloids have been pointed out by various investigators and cursorily reviewed in this journal.[1]

Erlanger and Hooker,[2] and later Hooker,[3] pointed out the relation of pulse pressure to renal secretion. More recently I[4] have studied the relation of pulse pressure to renal secretion in the intact kidneys of the dog, by modifying the pulse pressure normally existing in that animal. With the methods employed for changing the pulse pressure no appreciable change in the volume flow of blood through the kidneys was noted. Therefore, the changes in urinary secretion accompanying changes in pulse pressure were ascribed to some specific effect of pulsation itself.

In the experiments cited, pulsation had a marked effect upon the rate of secretion and upon the urea, sodium chloride, and albumin content of the urine. As a rule the rate of secretion and the urea and sodium chloride content of the urine were greater during the periods of pulsatile than during the periods of constant pressure. In a few experiments in which albumin appeared in the urine the amount secreted was greater during the periods of constant than pulsatile pressure.

Whether comparable results might not be produced under more artificial conditions seemed an interesting and important question.

[1] Gesell: This journal, 1913, xxxii, p. 70.

[2] Erlanger and Hooker: Johns Hopkins Hospital Reports, 1904, xii, p. 346.

[3] Hooker: This journal, 1910, xxvii, p. 24.

[4] Gesell: *loc. cit.*

Therefore, various solutions were filtered through different kinds of membranes in the hope that some light might be thrown upon the effect of pulsation upon secretion.

The apparatus employed is shown diagrammatically in figures 1, 2 and 3.

The high pressure filtration apparatus consists of pieces 1, 2, 3,

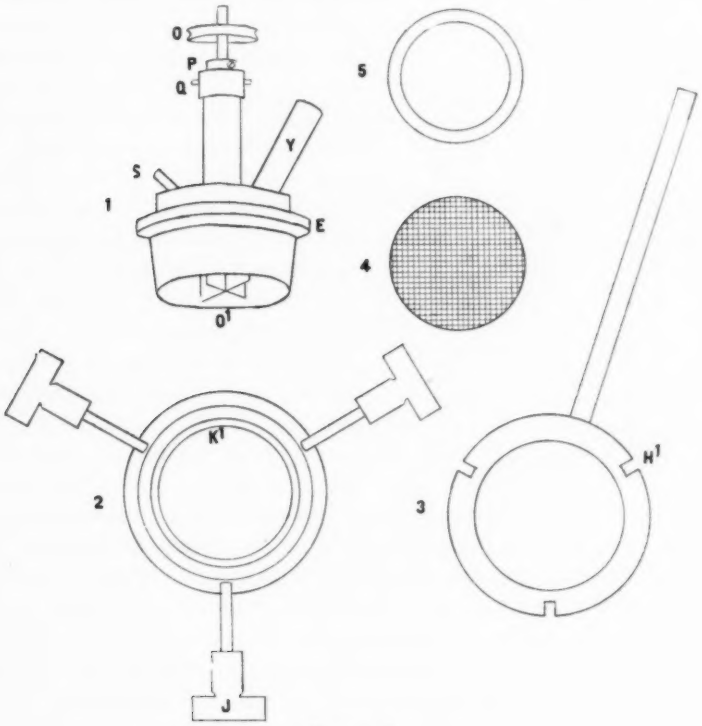

FIGURE 1

4 and 5 in Figure 1. See also Figure 3. The membrane employed is placed on number 5, a thin aluminum ring, and the whole placed upon a supporting perforated disc, number 4. The disc, in turn, is placed in number 2, where it rests securely upon ridge *K'*. Rubber washers are then placed over the membrane. Number 1 fits into number 2. Number 3 fits over ridge *E* of number

1, and adjustable arms *J* of number 2 fit into notches H^1. This arrangement serves to bring the circular edge of number 1 down securely on to the washers over the membrane. *O'* is a four-blade stirring wheel attached to a shaft passing through the packing box, *Q*, and is run at high speed by a motor belted with pulley *O*. *P* is a cuff fitted to the movable shaft; thus the distance between the stirrer and the membrane can be adjusted. *Y* is a large tube through which the pressure upon the membrane is transmitted. *S* is a smaller tube used for filling the apparatus with the filtrans and to record later the pressures prevailing in the filtrans.

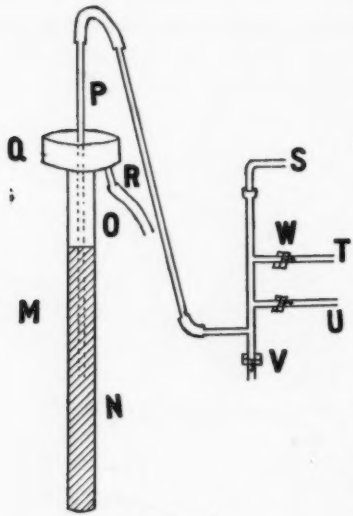

FIGURE 2

Pressures of 15–760 mm. Hg were employed. City water pressure was used as the source of pressure. In order to maintain this at a constant level the arrangement shown in Figure 2 was employed. *N* is a high glass tube containing mercury and *P* a graduated glass tube immersed in the mercury and connected by pressure tubing to hydrant, *S*. Pressure exerted through tubes *T* and *U* was regulated by simply raising or lowering tube *P*. The hydrant was then opened until a fairly brisk stream of water issued from the immersed end of tube *P*. The excess of water required to produce the desired pressure was drained from reservoir, *Q*, through tube *R*.

To convert the constant pressure prevailing in either tube *T* or *U* into a pulsatile pressure, the arrangement shown in Figure 3 was employed. The heavy filtering apparatus was put together as described. A glass cylinder *E* was firmly connected by a rubber stopper to the tube *Y*. The top of the cylinder contained another rubber stopper through which passed a stopcock, *A*, and the right-angle arm of a large calibre *T* tube, *B*. A soft rubber tissue bag containing a couple of drops of Hg was tied tightly about the

right-angle arm protruding into the glass cylinder. The tubes *K* and *F* were connected with the constant head of pressure. Into *K* was fitted a rotating stopcock, by means of which constant pressure was converted into pulsatile pressure. *F* is used to deliver constant pressure. *W* is a Hürthle manometer and *Z* a tube connected with the source of filtrans — both in connection with the filtering chamber through the tube *S*. The chamber was filled

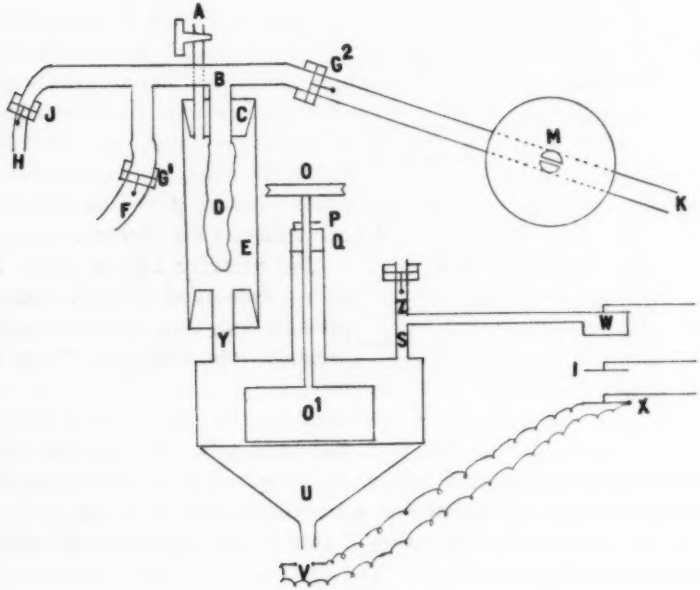

FIGURE 3

with filtrans by opening cocks *A* and *Z*. Cocks *A* and *Z* are closed when the cylinder, *E*, and the manometer system are filled to the exclusion of all air bubbles. The apparatus was then ready for filtration.

If constant pressure is desired clamp G^2 is closed and clamps G^1 and *J* opened and adjusted to produce any desired pressure. With G^1 open, the water rushes into the rubber bag, *D*, and exerts its pressure from the inside of the bag without mixing with the filtrans. As filtration proceeds the space made by lost filtrans is

occupied by water in the bag, and may occupy a large portion of glass chamber, *E*. The filtrans can then be renewed by momentarily closing clamp G^1 and expressing the water in the rubber bag by admitting fresh filtrans through *Z*.

If pulsatile pressure is required clamp G^1 is closed, G^2 opened, and the rotation stopcock, *M*, set in action by motor and pulley. Cock *M* is so constructed that communication between the filtrans and constant head of pressure is made and broken very abruptly so as to effectually produce sudden pressure changes. When cock *M* opens the pressure in *D*, and therefore in *E*, suddenly becomes approximately equal to the source of pressure in *K*. On closure of the stopcock the small amount of water which has accumulated in *D*, due to a certain amount of give in the apparatus, escapes through tube *H* and lowers the pressure in *E*. The clamp, *J*, regulates the amount of fluid escaping from tube *H* and therefore the fall in pressure during the period of closure of the stopcock, *M*. With tube, *H*, wide open systolic pressure can be made to rise to 760 mm. Hg and the diastolic pressure fall to zero. Gradually closing clamp, *J*, — diastolic pressure is rapidly raised, systolic pressure slowly raised, so that the pulse pressure gradually decreases to zero with the clamp entirely closed.

The rate of pulsation is regulated by the speed of the motor. If stirring is required a belt is slipped into pulley *O*. The pressure prevailing in the filtration chamber is recorded by means of the Hürthle manometer, *W*, the rate of filtration by drops, as it falls from the glass funnel *U*, onto the drop recorder *V*. *X*. Time is recorded in seconds by *I*.

Figure 4 is a record of pulsatile pressure produced by the method described.

In the experiments, alternating periods of constant and pulsatile pressure were used. At the outset, it is of importance to decide what constant pressure should be employed. If filtration is more rapid during pulsatile pressure than during constant mean pressure, pulsation might be considered as having some effect. The criticism might be raised that the greatest amount of filtration occurred only at systolic pressure, and therefore any experiment in which mean pressure was used as constant pressure would not be acceptable. But in the experiments performed the rate of

filtration did not change abruptly when a certain pressure was reached, but roughly varied with the magnitude of the pressure. During pulsatile pressure there is, with every pulsation, a long period of relatively low pressure, and a short period of relatively high pressure. During the long period, filtration is slower than during the short period. The mean pressure which divides these periods

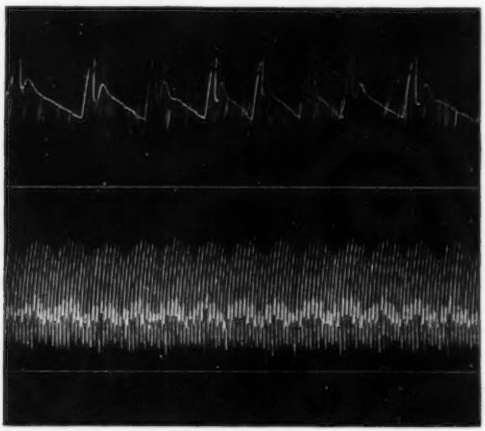

FIGURE 4

should, therefore, give a pressure which acting through the duration of a pulsation should be as effective as the combination of a period of low and high pressure prevailing in the pulsation, — provided pulsation itself has no effect upon filtration.

TABLE I

FILTRANS. — 2% NaCl.
MEMBRANE. — Heavy, coarse, loose mesh paper.
NO STIRRING.

Interval	Pulsatile or constant pressure	Mean pressure in cm. H_2O	No. of drops of filtrate per minute
5	P.P.	135	75
6	C.P.	135	32
7	P.P.	135	38
8	C.P.	135	23

Table I gives the results of an experiment in which a 2 per cent solution of sodium chloride was used as filtrans, and a piece

of heavy coarse loose mesh paper, 1 cm. in diameter, as membrane. Constant and mean pressure of the pulsatile pressure were each 135 cm. of H_2O.

During pulsatile pressure filtration was rapid, but fell to less than half the rate during the following period of constant pressure. On the return of pulsatile pressure filtration increased, not reaching the initial rate, but exceeding that of the preceding period of constant pressure. During the last period of constant pressure there was again a marked falling off of filtration.

An important point to be noted in this table, which also holds for most of the experiments performed, is that as the experiment progresses filtration gradually diminishes in rate — least, however, during the periods of pulsatile pressure. That is, during periods of pulsatile pressure the membrane tends to recover its original permeability.

TABLE II

FILTRANS. — Defibrinated dog's blood 400 c.c.
7% NaCl 1200 c.c.
MEMBRANE. — S.S. Gehärtete F.P. No. 575.
NO STIRRING.
PRESSURE. — Mean pressure (80 mm. Hg) throughout experiment.

Interval	Pulsatile or constant pressure	Relative rates of filtration	Per cent decrease in rate of filtration
1	P.P.	16	
2	C.P.	12	25
3	P.P.	10	16
4	C.P.	7	33
5	P.P.	6	14

Table II gives the results obtained on filtering a mixture of defibrinated blood and 0.7 per cent sodium chloride solution through hard filter paper. Constant and mean pressure were each 80 mm. Hg. The results, although not as marked, are comparable to those shown in Table I. Again, there was a gradual diminution in the rate of filtration throughout the experiment. In no case did the rate of filtration during a period of pulsatile pressure exceed that of the preceding period of constant pressure. Yet, if the percentage decrease during each period is calculated, it will be

noted that the greatest decrease occurred during the periods of constant pressure.

TABLE III

FILTRANS. — Defibrinated dog's blood 800 c.c.
Ringer's solution 1200 c.c.
Urea 10 g.
MEMBRANE. — Heavy, coarse, loose mesh paper as in 1.
NO STIRRING.
PRESSURE. — Mean pressure (80 mm. Hg) throughout experiment.

Interval	Pulsatile or constant pressure	No. of drops for each consecutive minute
1	P.P.	1.5, 1.5, 1.5, 1.5, 1.5, 1.5
2	C.P.	0.5, 0.5, 0.0
3	P.P.	2, 2, 2
4	C.P.	0, 0, 0, 0
5	P.P.	2, 2
6	C.P.	0, 0, 0, 0

Table III gives the results of filtering a mixture of Ringer's solution and defibrinated dog's blood through a loose mesh paper membrane. Constant and mean pressure were each 80 mm. Hg. The results in this experiment are the most striking obtained. During pulsatile pressure fairly rapid filtration occurred. During constant pressure little or no filtration occurred.

The question arises what is the cause for such marked changes in rate of filtration under the two conditions given.

At the close of every experiment in which a colloidal suspension was used as filtrans, a slimy, sometimes rather tenacious membrane was seen adhering to the upper surface of the filter. This slimy layer, which thickens as the experiment progresses, presumably adds more and more resistance to the passage of the filtrans through the membrane. Judging from the small amount of give to the apparatus with each pulsation, there should be little stirring of the filtrans at the membrane. Yet it seemed probable that the main effect of pulsatile pressure, so far observed, might be that of preventing the accumulation and concentration of colloids on the surface of the membrane. Therefore a few experiments were performed in which the filtrans was kept in active agitation by means of a rotary stirrer run at high speed. In these experi-

ments the formation of the slimy layer was considerably prevented. It was noted that the rate of filtration did not decrease as rapidly during the progress of the experiment as in the cases in which stirring was not employed.

TABLE IV

FILTRANS. — Egg white 25 c.c.
Ringer's solution 500 c.c.
MEMBRANE. — 1.5% celloidin membrane.
STIRRING.
PRESSURE. — Systolic pressure (40 cm. Hg) used as constant pressure.

Interval	Pulsatile or Constant Pressure	Stirring S or no stirring O	Drops of filtrate per minute
1	P.P.	S	8, 10, 10, 10, 9, 8, 8
2	C.P.	O	8, 5, 5, 6, 4
3	P.P.	S	5, 7, 10, 8, 9
4	P.P.	O	9, 7, 6, 6, 4, 5, 4, 4
5	P.P.	S	5, 6, 7
6	P.P.	O	7, 6, 4

Table IV gives the results of filtering a mixture of egg white and Ringer's solution through a 1.5% collodium membrane prepared according to the method of Bechhold.[1] Periods of constant and pulsatile pressure, with and without stirring, were employed. Systolic pressure of the pulsatile pressure and constant pressure were each 40 cm. Hg. The experiment shows the importance of stirring. With stirring the initial rate of filtration tended to be maintained for some time, while without stirring, even during periods of pulsatile pressure, the rate of filtration rapidly fell off. During period 2 of constant pressure, equal to systolic pressure of pulsatile pressure, filtration was not any faster than during pulsatile pressure, not even during the first minute, presumably before a membrane could have formed on the filter.

Table V gives results of an experiment similar to the preceding one. In this case a 1% membrane was used, and systolic pressure likewise employed as constant pressure. The results again show the beneficial effect of stirring, not alone during periods of constant, but also during periods of pulsatile pressure, although they are probably more marked during the periods of constant pressure.

[1] BECHHOLD: Zeitschrift für Physikalische Chemie, 1907, x, p. 257.

In number 3, during constant systolic pressure, plus stirring, filtration was not any faster than during the preceding period of pulsatile pressure, plus stirring. During period number 3 of constant pressure the rate of filtration gradually diminished, while in the preceding period of pulsatile pressure the rate increased.

TABLE V

FILTRANS. — Egg. white 25 c.c.
Ringer's solution 500 c.c.
MEMBRANE. — 1% glacial acetic collodium membrane.
STIRRING.
PRESSURE. — Systolic pressure (40 cm. Hg) used as constant pressure.

Interval	Pulsatile or constant pressure	Stirring S or no stirring O	Drops of filtrate per minute
1	P.P.	O	10, 11, 9, 8, 8, 7, 8, 6,
2	P.P.	S	7, 10, 11
3	C.P.	S	11, 10, 9, 10, 8, 9, 9, 10, 7, 7, 10, 10, 9
4	C.P.	O	9, 7, 6, 7, 6, 5, 4, 5, 5, 4, 5, 4, 4, 5, 3, 4, 4, 4, 4, 4, 4, 3, 4
5	C.P.	S	5, 6, 7, 7, 7, 7, 8, 7, 8, 6, 7, 5, 6, 7, 7

TABLE VI

FILTRANS. — Ringer's solution 1000 c.c.
Casein 30 g.
Phenolphthalein.
$Ca(OH)_2$ to slightly alkaline reaction.
MEMBRANE. — Dried peritoneum of the dog.
STIRRING.
PRESSURE. — Systolic 75 cm. Hg. Diastolic 70.
Constant pressure 73 cm. Hg.

Interval	Pulsatile or constant pressure	Drops per minute of filtrate
1	P.P.	56
2	C.P.	46
3	P.P.	48
4	C.P.	37
5	P.P.	44
6	C.P.	34
7	P.P.	33

Table VI gives results of filtering through the dried peritoneum of the dog, a mixture of calcium caseinate in Ringer's solution slightly alkaline to phenolphthalein. Alternating periods of constant and pulsatile pressure with constant stirring were employed. A pressure somewhat above mean pressure was used as constant pressure. As in most experiments there was a tendency of the rate of filtration to decrease with the progress of the experiment, but in this experiment that tendency was not very marked. In fact in two instances, periods 3 and 5, of pulsatile pressure the rate of filtration was more rapid than in the respective preceding periods of constant pressure.

Changes in the Nature of the Filtrate

A number of experiments were performed to determine whether there were any qualitative differences in the filtrate passing through the membrane during periods of constant and pulsatile pressure. In all of these experiments the rotary stirrer was run continuously at high speed, and alternating periods of constant and pulsatile pressure used as before. As constant pressures mean and diastolic pressure of the pulsatile pressure were employed.

Berlin blue, calcium caseinate, egg albumin, defibrinated dog's blood and milk, diluted with 0.9% sodium chloride or Ringer's solution, were filtered through collodium membranes and dog's peritoneum.

Such diffusible substances as urea and sodium chloride, with the methods employed for their detection in the filtrate, showed no quantitative relation to constant and pulsatile pressure.

Only in a few experiments did there seem to be a definite relation between the amount of colloids in the filtrate and the type of pressure employed during filtration. When a mixture of Berlin blue and a 1% sodium chloride solution was filtered through a collodium membrane there was no evident relation between the amount of blue in the filtrate and the type of filtration pressure employed.

Casein, in whole milk, diluted with Ringer's solution, filtered through a collodium membrane slightly permeable to casein, also showed no relation between the amount of casein in the filtrate and the type of filtration pressure employed.

The same results were obtained on filtering a 3% solution of calcium caseinate in Ringer's solution slightly alkaline to phenolphthalein, through the dried peritoneum of the dog. The filtrate on standing, however, showed a flocculent precipitation of casein. In the filtrate of pulsatile pressures the precipitation was coarser and occurred more rapidly. Furthermore, the reaction of the filtrans on passing through the membrane was evidently changed — for the filtrate from constant and pulsatile pressure was whiter than the filtrans, but less white from the periods of constant pressure than from the periods of pulsatile pressure. Nothing definite concerning these results can be stated. Further experiments are necessary.

Filtration of egg albumin through collodium membranes showed no quantitative relation to the type of filtration pressure employed. Neither did defibrinated dog's blood, diluted with Ringer's solution, when filtered through collodium membranes show any relation between the amount of albumin filtered and the type of filtration pressure.

In three experiments, however, in which a mixture of defibrinated dog's blood and Ringer's solution was filtered through the dried peritoneum of the dog, with the methods employed for the detection of globulin and albumin, there seemed to be a definite relation between the amount of globulin in the filtrate and the kind of pressure employed. Albumin was tested for and showed no relation to the type of pressure. The tests for globulin gave a cloudier solution with filtrates obtained during periods of constant than during periods of pulsatile pressure. Of these three experiments — diastolic pressure was used as the constant pressure in two, and mean pressure as constant pressure in the other. Even though higher pressures prevailed during the periods of pulsatile pressure, less globulin was forced through the membrane than during periods of relatively lower constant pressure.

Theoretical Considerations

Concerning the *causal* relationship of the changes in the nature of the filtrate accompanying changes from constant to pulsatile pressure, nothing definite can be offered.

The first thing that comes to mind is that either the pores in the membrane are larger during constant pressure than during pulsatile pressure, allowing the passage of colloidal particles through the membrane, or that the colloidal particles are larger during pulsatile pressure than during constant pressure, and therefore cannot pass through the membrane.

The first view does not seem tenable, for, if the pores of the membrane were larger during periods of constant pressure than during periods of pulsatile pressure, the rate of filtration should also be more rapid. This, however, was not the case.

The latter view would seem the more probable explanation: that is, the effect of pulsation may be due to the formation of molecular aggregates, too large to pass the pores of the membrane. Whether this formation of molecular aggregates would most likely occur at the surface membrane (junction between the filtrans and peritoneum) or throughout the filtrans, by agglutination of particles that may be brought into more intimate contact by pulsation, is hard to say.

In this regard the work of Ramsden is significant. Ramsden [1] studied the effect of shaking upon a solution of egg albumin. A clear solution, he found, becomes turbid with the production of fine coagulated strands of albumin which are no longer soluble in the medium. More recently [2] he has extended his experiments to other solutions and suspensions. He found, "that quite apart from evaporation, solid or highly viscous coatings are spontaneously and more or less rapidly formed upon the free surfaces of all proteid solutions; that similar coatings of solid or highly viscous matter occurs on the free surfaces of a large number of non-proteid colloid solutions and fine suspensions, and of a few apparently crystalloid solutions, and that they are formed also at the interfaces of solutions which without being of high viscosity are capable of persistent emulsion."

Ramsden found, "that by simple mechanical means adapted to produce heaping up of surface membranes, large masses of solids (mechanical surface aggregates) can be separated from all pro-

[1] RAMSDEN: Mann's Chemistry of Proteins, p. 275.

[2] RAMSDEN: Proceedings of the Royal Society of London, 1904, xxii.

teid solutions and from a large number of colloid solutions and suspensions."

Winkelblech [1] also demonstrated the formation of surface membranes between solutions of gelatin, egg albumin and other solutes and benzene.

Such surface membranes also form at the junction of a solid and a liquid. In the experiments cited in this paper the peritoneum might be considered the solid, and the blood the liquid. At the junction of the peritoneum and the blood there might be formed, by the concentration of colloids, a thick surface membrane. If no "means adapted to produce heaping up of surface membranes" are used, for instance, if constant pressure is employed for filtration, the formation of larger mechanical surface aggregates will not be encouraged. Therefore, if the particles on the membrane are smaller than the pores of the peritoneum, the colloid should be found in the filtrate.

Pulsation, however, is favorable to the formation of molecular aggregates, and, theoretically, should heap up the surface membrane with the formation of molecular aggregates, possibly large enough to prevent their passage through the peritoneum. This explanation of passage of colloids through the peritoneum requires changes in the filtrans only at the surface membrane, namely, the junction of filtrans and peritoneum, and therefore the process must not necessarily be reversible, or at least not quickly reversible.

Another possible explanation is that pulsation might favor the agglutination of the suspended colloidal particles, not only at the surface membrane, but throughout the filtrans. If this be the case, the process must be rather quickly reversible, for the difference in passage of colloids through the peritoneum during constant and pulsatile pressures can be demonstrated repeatedly in the use of one and the same sample of filtrans. So far the apparatus required for the investigation of this point has not been available.

Other less definite suggestions might be offered, but at present the formation of molecular aggregates seems most likely.

[1] WINKELBLECH: Zeitschrift für angewandte Chemie, 1906, p. 1953.

The Rate of Filtration during Constant and Pulsatile Pressure

In considering the effect of pulsation on the rate of filtration, there are three possible factors to keep in mind: 1. The size of the pores of the membrane; 2. The size of the colloidal particles to be filtered; and 3. The nature of the surface membrane, that is, the junction between the filtrans and filter. The possibly greater effect of a sudden impact in driving smaller particles through a membrane should also be kept in mind.

1. Size of pores. This may be considered from two aspects: (*a*) During pulsatile pressure the high pressure momentarily obtaining at the maximum level might stretch the membrane and so increase the size of the pores. This does not seem very probable for two reasons. One is, that rather thick and strong membranes were used, and they were supported on a perforated plate with circular holes one half mm. in diameter. It seems that a very great force would be necessary to stretch the membranes over such a small orifice. The part of the membrane supported is considerably greater than that unsupported. The compression of the supported membrane, therefore, might more than counteract any stretching over the unsupported areas.

Another thing which speaks against the stretching of the pores by the pressure obtaining during the height of the pulsatile pressure is that no more colloids are found in the filtrate during pulsatile pressure than constant pressure, indeed, in a few experiments, less have been found. It is possible, however, that there may be a balance between the amount of stretching of the pores and the relative increase in size of the molecular aggregates over that of the original suspended colloidal particles.

(*b*) During constant pressure the membrane may be compressed and, therefore, the size of its pores decreased.

Bechhold [1] points out the importance of the elasticity of the membrane for the difference in rate of filtration which he noted on filtration under constant pressure and slow intermittent pressure (not a true pulsatile pressure) in which the pressure was more or less slowly elevated and maintained for 15 minutes, and then again

[1] Bechhold: "Gedenkbock — Van Bemmelen," 1910.

released for the same time. Bechhold suggests that on increase of pressure, the membrane is compressed and the filtrans in the membrane is pressed to the farther side as filtrate. On the release of pressure the membrane due to its elasticity comes back to its normal size and like a sponge sucks itself full of filtrans again. According to Bechhold, intermittent pressure does not destroy the elasticity of the membrane nearly as much as a constant pressure. With the constant pressure the membrane is compressed and the pores necessarily diminished in size.

2. Simple stirring of filtrans. The formation of a rather tenacious membrane on the upper surface of the filter, and its slowing effect on the rate of filtration has been mentioned, also the prevention to a great extent of the formation of this membrane by the continuous use of the rotary stirrer. Stirring had a very marked effect upon filtration during periods of pulsatile as well as of constant pressure. Without stirring, pulsation had a very marked effect upon the rate of filtration and undoubtedly this was due largely to prevention of the accumulation of colloids at the filter. But even with vigorous stirring, pulsation seems to have an accelerating effect upon the rate of filtration. Whether this effect is simply additive to the effect of stirring — that is, produces more effective stirring, is hard to say. If the enhancing effect of pulsation is that of stirring, it is probably stirring of a different nature than that produced by the rotary stirrer, namely stirring by impact.

3. Impact. The velocity imparted to particles of the filtrans at the site of the membrane may be of some importance in helping their passage through the membrane during periods of pulsatile pressure. The fact that larger particles, as the colloids, which have a relatively high inertia, are found in different proportion in filtrates from periods of constant and pulsatile pressure, might support this view.

4. That pulsation might exert an enhancing effect by breaking up of the surface membrane (the junction of the filter and filtrans) which spans the pores or larger irregularities of the membrane is another possibility.

5. Keeping colloids in coarse suspension. Upon the assumption that pulsation favors the formation of molecular aggregates, either

at the filter or throughout the filtrans, the pulsation might favor filtration by keeping the colloids in coarse enough suspension to retard their entrance into the membrane and thereby diminish the clogging effects of these colloids.

Summary

A method is described for filtration with alternating periods of constant and pulsatile pressure.

Numerous solutions were filtered through a variety of membranes under constant and pulsatile pressure — with and without stirring, to determine whether pulsation had any effect: (1) Upon the rate of filtration; (2) upon the nature of the filtrate.

Rate of filtration. In experiments in which the rotary stirrer was not employed, pulsation favored filtration.

Stirring in itself had a very marked enhancing effect upon the rate of filtration, during periods of constant as well as pulsatile pressure.

With continuous stirring, filtration during periods of pulsatile pressure was more rapid than during periods of constant mean pressure.

In a few experiments in which constant stirring was employed, filtration during periods of a constant pressure at the systolic level was not any faster than during periods of pulsatile pressure.

Nature of the filtrate. With the methods for the detection of such diffusible substances as sodium chloride and urea — the amount found in the filtrate bore no relation to constant or pulsatile pressure.

With the methods used for the detection of colloids in the filtrate globulin (dog's defibrinated blood) alone seemed to show a relation to the type of filtration pressure employed — more globulin passing the membrane during periods of constant than during periods of pulsatile pressure.

CONCERNING THE PERIODIC CARDIOVASCULAR AND TEMPERATURE VARIATIONS IN WOMEN

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Historical Introduction.

THE theory that the life-processes in women follow a rhythmical wave-like movement, was suggested first by Dr. Mary Jacobi[1] in 1876. Her observations on the pulse, temperature, blood-pressure and muscular strength have been repeated and extended by a number of other workers and, in general, the conclusions have been that the highest point in all of these processes is reached from two to three days before the onset of the menstrual period, that they sink to the lowest point at its close, gradually rising to normal during the intermenstrual interval.

Much of the literature on this subject has been reviewed by Zuntz,[2] 1906, and by Hansen,[3] 1913, so that, with the exception of the observations on blood-pressure, I shall refer only briefly to it.

Variations in muscular strength during the menstrual period have been studied by Jacobi,[1] Ott,[4] Bossi,[5] Mandl and Bürger.[6] These have, in general, shown a decline corresponding to that of the other life-processes observed.

Although a number of metabolism experiments have been made on pregnant women, von Schroder's[7] furnishes the only reliable study of those menstruating. In three subjects he found a retention of nitrogen immediately before and during the menstrual period. It is an interesting fact that, in dogs, during the proœstrum, Murlin[8] has obtained a corresponding change. Blair Bell[9] records a diminution in the calcium content of the blood. Zuntz[2] states that the minimal exchange of energy is not altered, for the oxygen intake and the carbon dioxide output show no periodic variations.

Changes in temperature during menstruation have been more extensively observed than the changes in any of the other life-processes. Beginning with Retabeau's [10] work in 1870, investigations have followed by Jacobi,[1] Goodman,[11] Stephenson,[12] Reinl,[13] Ott,[4] Giles,[14] Vicarelli,[15] Mandl and Bürger,[6] Riebold,[16] Van de Velde,[17] Zuntz,[2] etc. The most thorough study is that of Hansen [3] in 1913, in which are given temperature curves not only for menstruating women but for normal pregnancy, confinement and lactation. In these a striking similarity is shown between the temperature curves of a man, of a girl before puberty and of a woman after the menopause as well as of women from whom the ovaries have been removed. Mandl and Bürger [6] have reported cases in which the wave form persists, though the removal of the uterus (the ovaries remaining intact) occasioned a cessation of the menses. Van de Velde [17] has observed the wave form in a case of vicarious menstruation. There seems then to be no doubt that there is a premenstrual rise in temperature, a menstrual and postmenstrual fall, followed by a return to normal, the average differences rarely amounting to more than a degree.

For respiration and pulse rate the curves follow those for temperature, but the wave form is less marked — according to Jacobi,[1] Ott,[4] Mandl and Bürger [6] and Zuntz.[2] Zuntz, who counted the pulse under minimum conditions, reports that during menstruation it is one to four beats below the premenstrual rate; the relation between the postmenstrual and the menstrual as well as that between the inter- and premenstrual was variable.

Blood-pressure observations were made by Jacobi,[1] at various intervals, on six normal subjects, through three menstrual periods, using Mohamed's sphygmograph. She does not indicate whether the tracings were taken under similar conditions. Her results show a minimum tension in the radial artery from one to four days after the cessation of the menses and a gradual return to a maximum seven to eight days before the next onset; occasionally, however, not until the first day of the flow. Stephenson,[12] reporting on four cases, found the arterial tension as measured by sphygmographic tracings — somewhat higher six to seven days before the menstrual period than on the day or two preceding. Ott,[4] with Basch's sphygmomanometer, obtained a fall in thirteen out

of fourteen cases; during the flow the pressure was almost constantly below the average, rising to normal again after the cessation. In 1897, Giles[14] studied seven patients admitted to the hospital for "trifling conditions," his records covering in all nine menstrual phases. Using the Dudgeon sphygmograph, he found the blood-pressure highest on the first two days of the period and on the day preceding, lower during the remainder of the epoch, rising again after the cessation. Wiessner[18] reported, in a brief note, observations made with the Riva-Rocci sphygmomanometer. He recorded the lowest pressure at the height of menstruation and a return to normal three to four days after the flow had ceased. The number of cases is not stated. Mosher,[19] using the Mosso instrument, recorded "under uniform conditions" the pressure in nine normal women. In some cases the records have extended over a period of forty-nine days. Most frequently she obtained a fall before the beginning of the menses, "the maximal fall being coincident with the onset," with a gradual return to normal by the time of cessation.

Mandl and Bürger,[6] 1904, studied two normal cases for fifty-one and fifty-three days. They employed the Gartner tonometer and found the average normal pressure to be from 110 to 120, rising to 150 immediately before the beginning of the menses and falling to 90 at the onset. In cases in which the uterus had been removed, they found the usual periodic rise and fall, while if the operation had included the ovaries also, the typical wave form was absent.

The most recent work, reported very briefly by Bogdanovics,[20] 1910, includes observations made with the Riva-Rocci sphygmomanometer, employing the Recklinghausen broad arm band. He obtained both the systolic and diastolic pressures and estimated the pulse pressure on two hundred and fifty persons, including some women who were pregnant and others who were ill. The number of normal women is not stated. In normal women, however, he reported a premenstrual rise of blood-pressure and a gradual decline after the onset of the menstruation. The maximum pressure fluctuated between 95 and 110 mm., the minimum varied from 36 to 52 mm. and the pulse pressure averaged 58 mm.

Observations and Discussion

My own observations, which I desire to report in this paper, were begun about two and a half years ago, in order to determine whether the periodic rhythm in blood-pressure is altered if regular exercise is taken during the menstrual phase as well as throughout the other phases of the cycle. It was necessary to secure daily records for each individual in order to be assured of the periodic

TABLE I

General Data Regarding Subjects

Subject	Age	Weight kilos	Height cm.	Days of record	Menstrual periods	Average duration of period
Group I						
A	18	59.1	162.5	96	4	3 days
B	17	59.5	165.0	84	3	3 "
C	18	54.88	162.5	93	3	5 "
Group II						
A	18	51.81	162.5	61	2	7 "
B	19	59.1	162.0	51	1	5 "
C	42	56.4	170.0	73	3	3 "
Group III						
A	36	52.27	160.0	79	3	3 "
B	40	63.6	170.0	37	2	3 "
C	32	48.6	166.7	(*a*) 63 (*b*) 57	3 2	4 "

rhythm. I was greatly surprised at being unable to discover, in the four subjects selected, the typical wave form in the blood-pressure curves so generally accepted. Thinking that possibly individual peculiarities were responsible for these negative results, the observations were continued on two other groups under different conditions but with practically the same results.

The subjects selected for this investigation were seven college students and four older women engaged in regular academic work. The ages ranged from 17 to 42 years. (Table I). All were in

normal health and all were in the habit of continuing their regular college duties during the menstrual phase. The college students frequently did not even interrupt their regular gymnasium exercise requirement of three hours weekly.

On the basis of the conditions under which observations were made, I shall separate the individuals into three groups. The

TABLE II

AVERAGES OF GROUP I

Phase	Systolic pressure		Diastolic pressure		Pulse pressure	
(*A*)	A.M.	P.M.	A.M.	P.M.	A.M.	P.M.
Intermenst.	111.54	108.9	75.42	75.65	37.9	36.9
Premenst.	109.2	111.69	73.42	74.5	35.33	35.19
Menstrual	111.34	111.4	74.2	76.72	36.16	34.0
Postmenst.	110.2	111.8	75.5	77.85	36.0	34.5
(*B*)						
Intermenst.	113.6	110.1	75.0	73.0	36.7	31.7
Premenst.	109.4	111.0	74.57	74.0	34.6	35.0
Menstrual	111.8	111.7	76.2	76.85	35.1	34.8
Postmenst.	113.3	113.3	72.66	75.6	39.0	36.6
(*C*)						
Intermenst.	111.2	114.0	71.97	77.2	37.4	35.8
Premenst.	109.8	115.8	72.0	73.5	36.0	41.2
Menstrual	113.4	114.0	74.6	77.5	45.5	36.5
Postmenst.	110.9	113.0	70.4	76.9	38.0	36.4

Erlanger sphygmomanometer with the standard cuff was employed and graphic records taken of both the systolic and diastolic pressures on the members of groups I and II. The systolic record was always checked by the auscultatory method, and whenever there was any uncertainty regarding the graphic record, that obtained by auscultation has been used.

Group I included four young women, who came to my office in the morning between 8 and 8:45 — half an hour to an hour

after breakfast, and in the afternoon between four and six. In the morning the building was reasonably quiet, in the afternoon there was, at times, a good deal of noise because of the proximity of the gymnasium. Every effort was made to secure identical conditions of quiet and repose in the subjects before observations were attempted. Since the records were continued

TABLE III

AVERAGES OF GROUP II

Phase	Systolic pressure		Diastolic Pressure		Pulse pressure		Temperature	
(*A*)	A.M.	P.M.	A.M.	P.M.	A.M.	P.M.	A.M.	P.M.
Intermenst.	112.6	114.0	76.15	81.6	34.8	32.4	97.7	98.32
Premenst.	112.75	114.85	73.5	79.7	37.75	34.6	98.41	98.02
Menstrual	112.0	117.4	78.6	82.4	33.2	33.0	98.26	98.5
Postmenst.	112.5	113.0	76.4	80.3	34.5	33.3	98.18	98.4
(*B*) Intermenst.	107.3	113.4	77.7	82.3	28.5	31.4	98.16	98.94
Premenst.	109.1	112.6	79.2	86.75	27.0	30.5	98.3	98.94
Menstrual	108.8	118.4	80.8	86.0	27.2	32.4	98.58	99.6
Postmenst.	106.0	110.0	78.0	81.0	27.5	29.0	98.3	98.6
(*C*) Intermenst.	99.35		72.85		28.0		98.1	
Premenst.	102.75		73.55		28.66		98.1	
Menstrual.	102.57		74.3		28.3		98.05	
Postmenst.	98.0		72.3		425.		98.1	

over long periods, and since the girls were familiar with the surrounding conditions and were accustomed to the noises, it was not felt that they were disturbing factors.

In order to have more constant conditions, however, the observations were undertaken on group II in a room of the infirmary located on the fifth floor of one of the college dormitories. This room was reached by an elevator, thus obviating disturbance due

to exercise. The subjects were four residents of this hall. The morning hour was from 8:15 to 9 and the evening 7:45 to 8:15.

In order to eliminate the possible interference of meals and to have conditions as nearly as possible at a minimum, the blood-pressure determinations were made on a third group of three before they arose in the morning, between six and seven o'clock.

TABLE IV

AVERAGES OF GROUP III

Phase	Systolic	Diastolic	Pulsepressure	Pulse rate	Temperature
(*A*) Intermenst.	96.02	64.27	28.15	63.23	
Premenst.	101.0	67.11	35.0	62.11	
Menstrual	95.27	65.65	29.63	59.63	
Postmenst.	95.8	68.2	27.6	59.36	
(*B*) Intermenst.	115.22	86.22	34.44	55.0	
Premenst.	112.62	80.75	31.87	57.7	
Menstrual	118.66	87.71	30.5	58.0	
Postmenst.	116.6	84.5	32.33	53.0	
(*C*) (*a*) Intermenst.	97.03	78.0	19.6	71.17	99.0
Premenst.	96.8	76.0	20.8	76.2	98.67
Menstrual	98.3	78.1	20.0	74.4	98.69
Postmenst.	101.0	79.3	21.66	75.8	98.9
(*C*) (*b*) Intermenst.	114.0	90.9	23.35	72.4	98.89
Premenst.	111.75	87.0	22.25	72.25	99.32
Menstrual	114.7	89.75	24.25	73.34	98.71
Postmenst.	114.5	91.14	23.6	70.7	98.29

Group III, *C* (*a*) averages of blood-pressure observations made with the Tycos sphygmomanometer, subject in reclining posture, between 6 and 7 A.M.

C (*b*) averages of blood-pressure observations made with the Erlanger sphygmomanometer, subject in sitting posture. Pulse rate and temperature taken with subject reclining, a few minutes before blood-pressures. Hour as in (*a*). Forty-nine days intervened between *a* and *b*.

The Tycos sphygmomanometer and the auscultatory method were used for both pressures. I checked this instrument frequently by a mercury manometer and found it constant and accurate. A

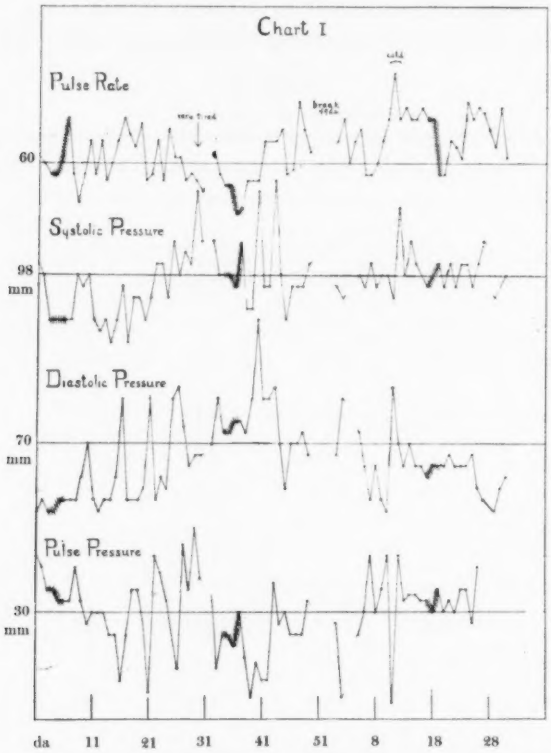

FIGURE I. Curves plotted from daily observations of pulse rate and blood-pressure made on *A* of group III, before she arose in the morning. The auscultation method with the Tycos sphygmomanometer was employed for the blood-pressure determinations. The menstrual periods are represented by the cross-hatched lines. See discussion on page 10, tables IV and VIII.

few weeks after these observations were discontinued, a second series was made under similar conditions on *A* and *C* of this group. On *A*, who was reclining, the Tycos was again used, but on *C*, who was sitting, the Erlanger instrument was employed.

Temperature records were obtained for groups I and II, taken by mouth, and for *C* of group III by rectum. The pulse was counted for one minute in groups I and III and in addition records of rectal temperature and of pulse rate were obtained, under minimum conditions, from two members of group I and for one of group III, the observations extending over periods of from 70 to 96 days.

The following results may be noted:

1. The temperature curves of groups I and II and the pulse curves of group I tend to follow the periodic rhythm indicated by numerous workers. This, however, is more obvious in those cases in which records were taken under minimum conditions (tables IV and Vb, chart II). The rectal temperature is, without doubt, more accurate than that taken by mouth. On one subject observations were likewise made on the rate of respiration and although these cover only forty days, in this case at least, there is an apparent rhythm.

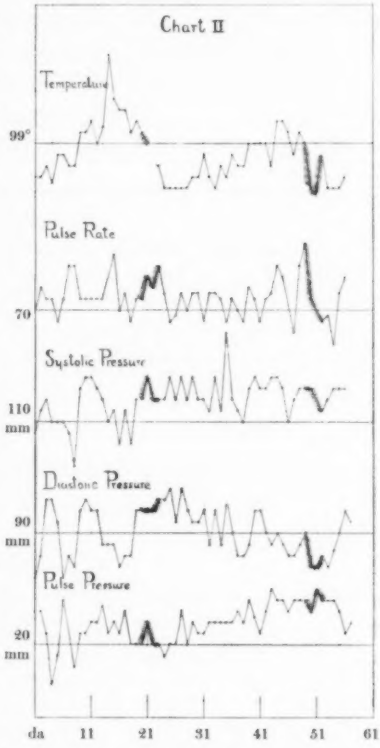

FIGURE 2. Curves plotted from daily observations of rectal temperature, pulse rate and blood-pressure made on *C* of group III. Temperature and pulse taken before the subject arose in the morning, blood-pressure determinations immediately afterward but with subject in sitting posture. Erlanger instrument employed. The menstrual periods are represented by the cross-hatched lines. See discussion on page 10, tables IV and VIII.

2. The blood-pressure determinations in the eleven cases studied have varied in duration from thirty-nine to ninety-six days. They have covered in all twenty-five complete menstrual epochs and thirty periods. The numbers given in the charts were obtained by averaging all

the records of the respective phases for each individual, thus securing a more accurate picture of the real values. The length of the pre- and postmenstrual phases, as I have taken them, depend upon the duration of the menstrual periods. That is, I have made the three periods equal and then considered the remaining days of the cycle as belonging to the intermenstrual interval. The data for two cases, I *D* and II *D*, are omitted since the former presented a marked case of amenorrhea and

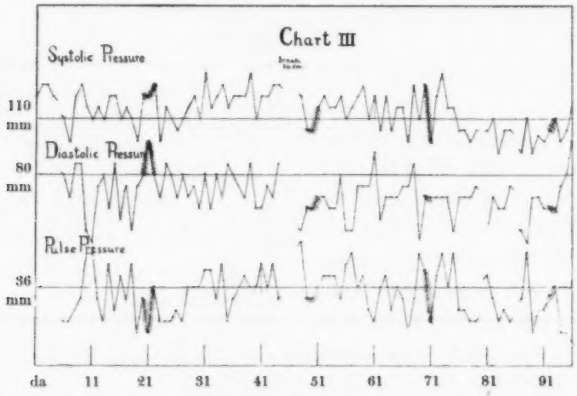

FIGURE 3. Curves plotted from daily observations of blood-pressure made on *A* of group I, between 4 and 6 P.M., subject in sitting posture. Erlanger instrument employed. The menstrual periods are represented by the cross-hatched lines. See discussion on page 10, tables II, V, and VI.

will be discussed in a later paper along with similar cases of irregular recurrences. In the record of II *D* there are a number of breaks so that it can hardly be considered a fair case. It should, however, be mentioned that neither gave any indication of a periodic rhythm.

With one exception the blood-pressure curves of all subjects fail to show a rhythmical movement. In fact as great variations may be observed during the intermenstrual phase as between the premenstrual and the menstrual. I feel confident that if the menstrual periods were not indicated on the charts (prepared from the records of each subject), one could not locate them with any degree of certainty.

In the exceptional case, *A* of group III (chart I, table IV) the systolic and pulse pressures undoubtedly show a rise two to three days before the onset of the period, then a gradual decline which continues throughout the menstrual and postmenstrual epochs. I desire to call special attention to the fact that the pressures at these times are not lower, sometimes not as low as those observed

TABLE V

GROUP I, C

(a) *Averages of mouth temperature and pulse rate taken at the same time as the blood-pressure records of chart I*

Phase	Temperature	Pulse rate	Menst. record above or below, Temperature	Pulse rate
Intermenst.	98.65	74.1	− 0.14	− 1.8
Premenst.	98.7	75.28	− 0.19	− 2.98
Menstrual	98.51	72.3		
Postmenst.	98.63	70.93	− 0.12	1.37

(b) *Averages of rectal temperature and of pulse rate taken between 6 and 7* A.M., *before rising 18 mo. later*

Phase	Temperature	Pulse rate	Menstrual records above or below Temperature	Pulse
Intermenst.	98.1	69.9	0.15	− 0 6
Premenst.	98.58	70.5	− 0.33	− 1.2
Menstrual	98.25	69.3		
Postmenst.	97.92	66.75	− 0.33	2.55

here and there during the intermenstrual days. The diastolic pressures show irregularities too great to permit an analysis of the curve. The two exceptional pressures, for reasons indicated on the chart, have been omitted from the averages. I am unable to account for the other striking variations.

C of group III (chart II, table IV) shows, undoubtedly, a periodic rhythm in the temperature variations and possibly in the pulse rate, but no such periodicity can be detected in the blood-pressure curves.

In *A* of group I the objection may be raised that conditions were not sufficiently constant to give the normal record in blood-pressure, but on comparing pulse and temperature records taken at this time (table V) with others taken in the morning before

TABLE VI

GROUP I. MENSTRUAL RECORDS ABOVE OF BELOW THOSE OF OTHER PHASES

Phase	Systolic pressure		Diastolic pressure		Pulse pressure	
(*A*)	A.M.	P.M.	A.M.	P.M.	A.M.	P.M.
Intermenst.	− 0.2	2.5	− 1.22	1.1	− 1.7	− 2.9
Premenst.	2.14	− 0.3	0.8	2.2	0.83	− 1.2
Postmenst.	1.14	− 0.4	− 1.3	− 1.13	0.2	− 0.5
(*B*)						
Intermenst.	− 1.8	1.6	1.2	3.85	− 1.6	− 3.2
Premenst.	2.4	0.7	1.6	2.85	0.54	− 0.15
Postmenst.	− 1.5	− 1.6	3.54	1.25	− 3.9	− 1.7
(*C*)						
Intermenst.	2.25	0	2.6	0.4	8.13	0.75
Premenst.	3.61	− 1.8	2.57	4.0	9.5	− 4.7
Postmenst.	2.54	1.0	4.17	0.6	7.5	0.1

arising, it may be seen that there are but slight variations in the differences in the two sets of records. Such a comparison would lead one to the conclusion that here the outward conditions were not important modifying factors. It was found more satisfactory to chart the morning and afternoon records separately, in the cases in which both were taken, as well as to prepare the two sets of averages. The blood-pressure curves inserted (chart III) were prepared from the afternoon determinations but are not significantly different from the morning curves.

Reviewing the entire series of observations, including those already discussed, it is evident that there are menstrual epochs in which all three pressures fell below the intermenstrual pressure, but the average differences are slight and on the other hand there are epochs in which they rose above the normal. If the averages for each phase be examined (tables II, III, IV) it will be seen that

TABLE VII

GROUP II. MENSTRUAL RECORDS ABOVE OR BELOW THOSE OF OTHER PHASES

Phase	Systolic pressure		Diastolic pressure		Pulse pressure		Temperature	
(*A*)	A.M.	P.M.	A.M.	P.M.	A.M.	P.M.	A.M.	P.M
Intermenst.	− 0.57	3.4	2.4	0.8	− 1.54	− 0.64	0.56	0.18
Premenst.	− 0.75	2.55	5.1	2.7	− 4.5	− 1.57	− 0.15	0.48
Postmenst.	− 0.5	4.4	2.13	2.1	− 1.3	− 0.33	0.1	0.1
(*B*)								
Intermenst.	1.5	5.0	3.1	3.66	− 1.3	1.0	0.42	0.66
Premenst.	− 0.3	5.8	0.6	− 0.75	0.2	1.9	0.28	0.66
Postmenst.	2.8	8.4	2.8	5.0	− 0.3	3.4	0.28	1.0
(*C*)								
Intermenst.	3.2		1.43		0.3		− 0.05	
Premenst.	− 0.18		0.73		− 0.4		− 0.05	
Postmenst.	4.57		2.0		2.9		− 0.05	

there is a tendency toward a premenstrual rise and a postmenstrual fall, but these differences are not great (tables VI, VII, VIII). The premenstrual rise occurred in the systolic pressures of six individuals, the greatest increase amounting to 5.73 mm. (chart I, table VIII.) The diastolic premenstrual pressure exceeded that of the menstrual in but two cases and in these less than 2 mm. (table VII, VIII). Since the pulse pressure is recognized as being of especial value as an indication of the efficiency of the heart, one might expect to find a regular variation here, yet the highest average difference in eight individuals was but 5.37 mm. (table VIII). The postmenstrual averages in about half of the subjects were a little below the menstrual and in the majority of

cases 1 to 2 mm. lower than the intermenstrual. If the product of the pulse pressure by the pulse rate be considered, it is found to be remarkably constant for each individual.

It is difficult to compare my blood-pressure findings with those of other workers, since they have not tabulated their results and only two have plotted them as curves (Stephenson and Mandl

TABLE VIII

GROUP III. MENSTRUAL RECORDS ABOVE OR BELOW THOSE OF OTHER PHASES

Phase	Systolic	Diastolic	Pulsepressure	Pulse rate	Temperature (rectal)
(*A*)	A.M.	A.M.	A.M.	A.M.	A.M.
Intermenst.	− 0.75	1.38	1.48	− 3.6	
Premenst.	− 5.73	− 1.56	− 5.37	− 3.48	
Postmenst.	− 0.53	− 2.55	2.03	0.3	
(*B*)					
Intermenst.	3.44	1.5	− 3.94	3.0	
Premenst.	6.04	6.96	− 1.4	0.3	
Postmenst.	2.0	3.2	− 1.8	5.0	
(*C*) (*a*)					
Intermenst.	1.3	0.1	0.4	3.23	− 0.31
Premenst.	1.5	2.1	− 0.8	− 1.8	0.02
Postmenst.	− 2.7	− 1.2	− 1.66	− 1.4	− 0.21
(*C*) (*b*)					
Intermenst.	0.7	− 1.15	0.9	0.94	− 0.18
Premenst.	2.95	2.75	2.0	1.1	− 0.61
Postmenst.	0.2	− 1.39	0.65	2.64	0.43

and Bürger). Faught[21] has brought out the difficulty of making comparisons between the figures obtained with the instruments used some years ago and those now in use, especially when the width of the cuff is not stated, or when it is not known whether any cuff was used.

I am aware that there are many possibilities of error in this work and that conclusions cannot be drawn from observations on

eleven subjects. Yet it is of great interest that records taken over long periods, with the most approved of modern sphygmomanometers, give little support to the results hitherto generally accepted.

Various theories have been brought forward to explain the diminution in temperature which undoubtedly occurs. Zuntz [2] concludes that, since his experiments indicate no change in heat production, there must be an increase in the heat given out. He suggests that this may be due to a change in the innervation of the blood-vessels; also that the profuse sweating, which he says occurs in many women during menstruation, may play a rôle in the temperature reduction. The results of my blood-pressure observations do not give much support to the innervation hypothesis, while in regard to the profuse sweating, such a condition did not occur in any of my subjects and seems to be a phenomenon unknown or unnoticed by normal women. According to Hansen,[3] the reduction may be explained on the basis of the decrease in protein metabolism. Dr. F. G. Benedict, of the nutrition laboratory (in a letter to the writer), regards a constantly lower pulse, when taken under minimum conditions, as a clearer proof of lower heat production than a mere falling temperature. One might regard the diminished pulse rate, where it occurs, as of greater significance than the fall in temperature.

Granted that lowering in pulse rate, temperature, and blood-pressure occur during the menstrual and postmenstrual periods, should much significance be attached to a diminution in pulse rate of from two to three beats, to temperature variations rarely greater than one degree, and to blood-pressure changes averaging from two to five millimeters? Certainly one would suppose that in a healthy individual a normal periodic function should not be accompanied by a marked depression in all of the life-processes and a generally lowered efficiency so emphasized by numerous writers. If there is a compensation for the loss of blood by the retention of nitrogen (Murlin),[8] then it would seem that there should be a compensation in the cardiovascular system and this is indicated in the tendency of the blood-pressure to show such slight variations.

SUMMARY

In the study reported, observations made on the pulse and temperature of women support the usually accepted theory of a rhythmical movement in the life-processes; the highest point is reached from three to four days before the menses, the lowest point about three days after their cessation. With one exception, blood-pressure records of the systolic, diastolic and pulse pressures made on eleven women gave such irregular results that they cannot be regarded as supporting the wave-theory.

My results, as far as they go, seem to indicate that there has been a tendency to overemphasize the inefficiency of women during the menstrual period.

It is a pleasure to express my appreciation of the encouragement and helpful suggestions received, during the progress of this work, from Dr. Lilian Welsh and from Dr. Donald R. Hooker. I wish also to thank the latter for the use of one of the Erlanger sphygmomanometers from his laboratory.

I am greatly indebted to my students and to the other women, subjects of the observations, for their intelligent interest and hearty coöperation in the work. One of the group, Miss Marion Janney, has prepared the charts.

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THE INFLUENCE OF CURARE ON VASOMOTOR REFLEX THRESHOLDS

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IN a recent communication from this laboratory dealing with the thresholds for certain vasomotor reflexes,[1] the point was made that in the ordinary experimental use of faradic stimuli shocks of undue intensity are frequently employed, particularly with curarized animals, in which obvious signs of excessive stimulation are not afforded. In connection with a discussion of the extent to which the use of very strong stimuli is justified the question was raised (p. 225) as to whether curare might not have so depressing an influence on vasomotor activity as to require powerful stimulation to overcome it. The present paper is a report of the results of our studies of this point.

The admirable investigations of Sollmann and Pilcher [2] have shown that *qualitatively* the vasomotor mechanism is not seriously modified by ordinary doses of curare, except for a transient peripheral block. These authors did not consider the question of a possible *quantitative* effect of the drug, since they employed maximal stimuli throughout their work.

Method. — Our experiments were performed upon cats, narcotized with ether. In these experiments we used continuous etherization, carried on thus: two ordinary lamp wicks were inserted in a small bottle of ether with about 5 cm. of each projecting. This bottle was then placed within a larger, wide-mouthed bottle provided with inlet and outlet tubes as in the ordinary ether bottle. By adjusting the exposure of wick a rate of ether evaporation just sufficient to maintain the desired degree of anaesthetization was readily obtained. With this method we had very uniform results.

[1] Martin and Lacey: This journal, 1914, xxxiii, p. 212.

[2] Sollman and Pilcher: This journal, 1910, xxvi, p. 233.

We administered curare by injection into the femoral vein. Our etherized cats were quite resistant to the drug. Thirty mg. per kilo body weight were regularly used, and often did not induce complete irresponsiveness to strong peripheral sciatic stimulation, although spontaneous breathing was abolished. For artificial respiration we used an air blast interrupted by a motor-driven valve.

We observed, as did Sollmann and Pilcher,[1] a marked and immediate fall of blood pressure following the injection of curare. Perhaps on account of our heavier doses, or because we used cats instead of dogs, the blood pressure was slower in returning to a persistent level than in their experiments. We also noted a difference from their results in that the persistent level after curare was lower than the original level, whereas in their animals it was usually higher.

We investigated the effect of curare upon the thresholds for blood-pressure drop and blood-pressure rise from central stimulation of the sciatic and saphenous nerves in the hind leg and of various branches of the brachial nerve in the front leg. We studied also the effect of the same drug on the thresholds for mild depression and profound depression from central stimulation of the combined vago-depressor trunk.[2]

Thresholds were measured in *Z* units.[3] The rate of stimulation varied between 8 and 15 per second.

Results. — Table I contains a summary of our results with sensory nerves other than the vagus. This table indicates a moderate lowering of the sensitiveness of the vasoconstrictor centre by curare, but not in our opinion a sufficient lowering either to invalidate the use of curare in vasomotor experiments, nor to justify the use of stimuli of supraphysiological intensity.

The experiments on central stimulation of the vago-depressor resulted as follows: the threshold for mild depression without curare (9 experiments) averaged 10 *Z* units; with curare (8 ex-

[1] SOLLMAN and PILCHER: *loc. cit.*, p. 239.

[2] MARTIN and STILES: This journal, 1914, xxxiii, p. xxxvi; and xxxiv, p. 106.

[3] MARTIN: The measurement of induction shocks, New York, 1912, p. 73. Also MARTIN and LACEY: *loc. cit.*

periments) 13 *Z* units. The threshold for profound depression (more than 24 per cent) without curare averaged for 10 experiments 245 *Z* units; with curare (9 experiments) 240 *Z* units. These results, even more clearly than those with other sensory nerves, show how slight is the influence of curare on the vasomotor centres. Although there appears to be no change of the threshold for profound depression by curare, certain facts that have come out in connection with this study suggest that the drug may have a definite effect on the vasoconstrictor centre.

TABLE I

THE INFLUENCE OF CURARE ON THE THRESHOLDS FOR REFLEX BLOOD-PRESSURE CHANGE. Z UNITS

	Thresholds for pressure drop				Thresholds for pressure rise			
Nerve Stimulated	No. expts. averaged	Threshold before curare	No. expts. averaged	Threshold after curare	No. expts. averaged	Threshold before curare	No. expts. averaged	Threshold after curare
Sciatic	7	14 *Z*	4	27 *Z*	5	168 *Z*	6	256 *Z*
Saphenous	5	5	4	16	2[1]	265	3[1]	500
Branch of Brachial	5	7	5	28	4	400	6	480

[1] We have on five occasions with and without curare failed to obtain a rise in blood pressure from central stimulation of the saphenous nerve with shocks exceeding 2000 *Z* units. Well-marked pressure drops were brought about in these cases by all the stimulations we administered.

In our paper on the two types of reflex blood pressure drop[1] the "all or none" character of the mild depression and the rather abrupt change to the profound type were mentioned. In comparing our series of graded stimulations with and without curare we find that in the curarized animals the change from the mild to the profound drop was apparently less abrupt than in the cases where no curare was used. In other words, the extent of pressure drop appears to bear a closer relation to the stimulation strength in curarized animals than in those that are uncurarized.

In the paper cited above we have described characteristic

[1] MARTIN and STILES: *loc. cit.*, p. 110

qualitative differences between the mild and profound types of pressure-drop.[1] These were first noted by Bayliss.[2]

Quantitatively we find that the mild type in uncurarized animals is rarely associated with an extent of drop exceeding 14 or 15 per cent, and that the profound type is rarely less than 20 to 24 per cent. In twelve experiments without curare including 97 mild depressions and 25 profound ones, there were only seven whose extent was between 14 and 20 per cent, whereas in nine experiments with curare, including 59 mild depressions and 25 profound ones, there were 13 with extent between 14 and 20 per cent.

TABLE II

EXPERIMENT OF NOVEMBER 25, 1913

THE GRADATION OF REFLEX BLOOD-PRESSURE DROP IN CURARIZED ANIMALS CONTRASTED WITH THE ABRUPT CHANGE OF CHARACTER FROM MILD TO PROFOUND PRESSURE DROP IN UNCURARIZED ANIMALS FROM CENTRAL STIMULATION OF THE VAGUS NERVE

Without curare		With curare	
Stimulus *Z* units	Percentile pressure drop	Stimulus *Z* units	Percentile pressure drop
1.87	7.1	1.87	10.2
3.25	9.2	22	16.8
5	10.3	45.5	18.9
22	10.8	60	25.7
35	12.3	73.2	32
60	9.2	127	38.5
127	12.3		
192	31		

This tendency toward an increased gradation of response after curarization can be illustrated also by the data from a particular experiment. Table II presents the record obtained on November 25, 1913. The table shows that in the uncurarized animal after

[1] MARTIN and STILES: *loc. cit.*, p. 110..

[2] BAYLISS: Journal of physiology, 1893, xiv, p. 314.

the full, mild effect was reached at 5 *Z* units a 25-fold multiplication of the stimulus to 127 units brought about no noteworthy increase in the extent of response, and that after that point was passed a 50 per cent increase in stimulation strength caused a response two and one-half times as great as the last one, this marking the region of transition from the mild to the profound type of depression. After the administration of curare, on the other hand, the response shows a definite tendency to increase as the strength of stimulus increases. A possible explanation of this effect of curare may be that it impairs the coherence of the vasoconstrictor centre, causing it to respond to sensory stimulation more or less piecemeal instead of as a unit. In the particular experiment cited in Table II there appeared likewise a definite increase of sensitiveness after curare. Our experience as a whole does not suggest that this latter is a necessary or characteristic effect.

Conclusion. — Our results may be summarized in the statements (1) that the thresholds for vasomotor reflexes are not, as a rule, markedly affected by curare; and (2) that the vasoconstrictor centre shows signs of impairment of unity under the influence of the drug, causing it under certain circumstances to exhibit gradations of response not usual in uncurarized animals.

FACTORS AFFECTING THE COAGULATION TIME OF BLOOD[1]

I. The Graphic Method of Recording Coagulation Used in These Experiments

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MANY methods have been devised for determining coagulation time. The description of these methods is unnecessary in this paper, for they have been considered critically in two comparatively recent reviews, one by Addis,[2] the other by Morawitz.[3] With different methods the coagulation time of blood (at 20°) has been set down as ranging from approximately 5 minutes (Addis) to 20 minutes (Morawitz and Bierich). This great discrepancy shows that there is no definite "coagulation time" quite independent of the method used, i.e., the conditions peculiar to any coagulometer are likely to affect the time of clotting. Since to drawn blood any instrument is a foreign body, all that is required of an instrument is that the conditions of its use shall be constant.

The conditions defined by Addis as being essential for accurate estimation of coagulation time are as follows:

1. The blood must always be obtained under the same conditions.

2. Estimates must all be made at the same temperature.

3. The blood must always come in contact with the same amount and kind of foreign material.

[1] A preliminary report of these experiments was presented at the meeting of the American Physiological Society, Dec. 29, 1913. See Proceedings, This journal, 1914, xxxiii, p. xxxviii; also p. 372.

[2] Addis: Quarterly journal of experimental physiology, 1908, i, p. 305.

[3] Morawitz: Abderhalden's Handbuch der biochemischen Arbeitsmethoden, 1911, v, pp. 235–252.

4. The end point must be clear and definite and must always indicate the same degree of coagulation.[1]

Besides conforming to these four conditions it seemed to us that the ideal instrument should also yield a permanent objective record, made by the blood itself. The form of coagulometer finally employed is illustrated diagrammatically in Figure 1. It consists essentially of a light aluminum lever[2] with the long arm nearly counterpoised by a weight *W*. The long arm is prevented from falling by a support *S*, and is prevented from rising by a horizontal

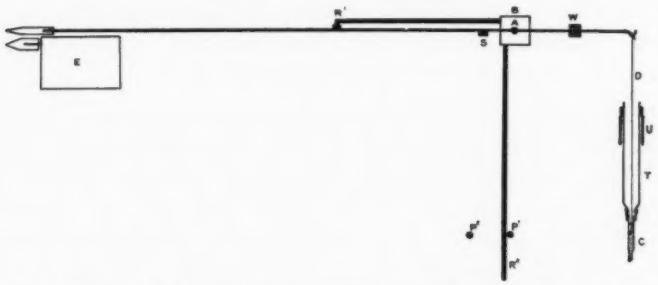

FIGURE 1. Diagram of the graphic coagulometer. The cannula at the right rested in a water bath not shown in this diagram. For further description see text.

right-angled rod reaching over the lever at R^1 and fixed into the block *B* which turns on the axis *A*. Into the same block is fixed the vertical rod R^2. When this rod is moved from the post P^1, against which it is held by the weight of the horizontal rod R^1, towards the other post P^2, the check on the long arm of the lever is lifted, and, if the short arm is heavier, the long arm will then rise.

The cannula *C*, into which the blood is received, is 2 cm. in total length and slightly more than 2 mm. in internal diameter. It is attached by a short piece of rubber tubing to the tapered glass tube *T*, 5 cm. long and 5 mm. in internal diameter. The upper end of this tube is surrounded by another piece of rubber which supports the tube when it is slid into the **U**-shaped support **U**, fixed directly below the end of the short arm of the lever.

[1] ADDIS: *loc. cit.*, p. 314.

[2] The "heart lever" made by the Harvard Apparatus Company.

By drawing the cannulas from a single piece of glass tubing and by making the distance from shoulder to upper end about 12 mm., receptacles of fairly uniform capacity are assured. All the dimensions, the reach of the rubber connection over the top of the cannula (2–3 mm.), the distance of the upper rubber ring from the lower end of the glass chamber (4 cm.), etc. — were as nearly standard as possible.

A copper wire *D*, 8 cm. long and 0.6 mm. in diameter, bent above into a hook, and below into a small ring slightly less than 2 mm. in diameter, is hung in a depression at the end of the short arm of the lever. The small ring then rests in the upper part of the cannula (see Figure 1). The weight of the copper wire makes the short arm of the lever heavier than the long arm by 30 mgm., when the delicate writing point is moving over a lightly smoked drum. Half a dozen of these standard wires were needed.

The precautions taken to fulfill the conditions stated above as essential for accurate determination of the coagulation time, were as follows:

1. Drawing the blood. — The blood was taken from the femoral artery. The artery (usually the right) was laid bare in the groin and freed from surrounding tissue. A narrow artery clip with each limb enclosed in soft rubber tubing, and with its spring exerting gentle pressure, was placed on the artery immediately below the deep femoral branch, thus allowing no blood to stagnate above the clip. Between the clip and a ligature applied about 1.5 cm. below, an opening was made. The blood was carefully milked out of the vessel between a blunt dissector moved beneath, and a small forceps, twisted into a pinch of absorbent cotton, moved above.

The cannula, cleaned in water, alcohol, and ether, was set in the rubber connection of the glass tube; the point of the cannula was then lubricated with vaseline, and slipped into the artery. The pressure of the clip on the artery was next very slightly released and blood was allowed to flow into the cannula up to the lower border of the rubber connection. Only a good-sized drop of blood was needed. Sometimes the blood ran one or two millimeters above or below, but without appreciably changing the result. Since the clip was situated on the femoral immediately

below a branch in which the circulation persisted, *the blood received in the cannula was always fresh from the moving stream.* As soon as the clip gripped the artery again, the cannula was slipped out. A helper then promptly milked the vessel in the manner described above, and covered it with a pad of absorbent cotton, smeared with vaseline, to prevent drying. Thereby blood was not permitted to stagnate; and when a new sample was to be taken, the vessel was clean and ready for use.

The tip of the cannula was at once plugged by plunging it into a flat mound of plasticine about 3 mm. high. It was drawn off sidewise lest the plasticine plug be pulled out again. One of the copper wires *D* was now slid into the tube and cannula, the tube slipped into the **U**-support, and the wire lifted and hung on the lever. This procedure, from the moment blood began to flow until the wire was hung, consumed usually about 20 seconds.

2. Uniform temperature.—Under the **U**-support was placed a large water bath, in which the cannula and the tapering part of the tube were submerged. A thermometer was fixed to the **U**-support so that the bulb came near the cannula in the bath. The water was kept within a degree of 25° C. This temperature was chosen for several reasons: (*a*) The cannula has room temperature and rapidly cools the small volume of blood that enters it. To heat blood and cannula to body temperature would take time. A bath near room temperature, therefore, seems preferable to one near body temperature. (*b*) The test of clotting was conveniently made at intervals of a half-minute, and if the clotting process were hastened by higher temperatures, this interval would become relatively less exact. (*c*) A temperature of 25° C. rather than lower was selected because, as Dale and Laidlaw [1] have shown, the coagulation time is much slower for a given change in temperature below 25° than for the same change above. And with slowing of the process the end point, when the determination depends on supporting a weight, is less likely to be sharp. (*d*) The researches undertaken with use of this coagulometer were concerned with factors hastening the process. For that reason and for reason (*b*),

[1] DALE and LAIDLAW: Journal of pathology and bacteriology, 1912, xvi. p. 359.

a long rather than a short coagulation time for normal conditions was desirable.

3. Uniformity in the amount and kind of contact with foreign surface. — The capacity of the cannulas was fairly uniform, as stated above; the amount received in them was fairly constant; and the wire hanging in the blood presented approximately the same surface in different observations.

A further condition for insuring consistent treatment of the blood in different cases was that of making the tests for coagulation always at the same intervals. Below the writing point of the lever was set an electromagnetic signal E which recorded half-minutes. At the moment a record was made by the signal (see first signal mark, Figure 2), the clip on the artery was opened, the blood taken, and the process thus begun. In about 20 seconds the cannula was suspended in the water bath, and the wire was hanging on the lever. At the next record by the signal and at every subsequent record the vertical rod R^2 was pushed with the index finger from post P^1 to post P^2 and allowed to move back.[1] This motion was uniform and lasted about one second. The check R^1 on the long arm of the lever was thus raised, and as the wire sank in the blood the writing point rose, recording that coagulation had not taken place (see Figure 2).

4. Definite end-point. — As soon as the blood clotted, the weight of 30 mgm. was supported, and the failure of the lever to rise to the former height in the regular time allowed recorded that the change had occurred.

Very rarely the swing of the lever would be checked for a moment and would then begin to move rapidly, indicating that a strand of fibrin had formed but not sufficiently strong to support the weight, and that when the strand broke, the weight quickly sank in the blood. When this occurred the next record almost always was the short line, which signified that the weight was well supported.

A very slight strand of fibrin was able to prevent the weight

[1] By applying a T-shaped wire to the axis of the second hand of a clock, and bending outward at right angles the ends of the top of the T, it is possible to have the vertical rod R^2 shifted automatically at half-minute intervals. This method was not used extensively in our experiments.

FIGURE 2. Record (reduced one-third) of five successive tests of coagulation, with the animal in a uniform condition. The lower line records intervals of 30 seconds. The marks below the time record indicate the moments when the blood samples were drawn.

from dropping, though at different times the amount of support differed, as shown by the varying length of the final lines (cf. first and last series, Figure 2). These variations are probably a rough indication of the degree of coagulation. In our experiments, however, the length of the final line was disregarded, and merely the fact that the lever failed to swing through its usual distance was taken as evidence of a clot, and the consequent short record was taken as the end point.

As soon as this end point was registered, the tube, wire and cannula were lifted out of the bath; the cannula was then separated from the tube and pulled away from the wire. The clot was thus disclosed, confirming the graphic record.

The method, at least when used at half-minute intervals, did not reveal in all instances the same degree of clotting. Usually, when the process was very rapid, the revealed clot was a thick jelly; whereas, when the process was slow, a strand of fibrin or at most a small amount of jelly was found. This difference in the *degree* of coagulation introduced, of course, an element of inexactness. In our experiments, however, this inexactness was unfavorable to the result we were seeking for, i.e., the acceleration of the process — because the jelly is a later stage than the fibrin strand; and since we nevertheless obtained good evidence of acceleration, we did not in these experiments attempt to determine more accurately differences in the stage of the clotting process.

Cleaning of apparatus. — After the wire was removed from the tube, the clot attached to its ring-tip was carefully brushed away

under cool running water. Under the running water, also, a trimmed feather was introduced into the cannula and the tube to push out the plasticine and to wash out the blood. Wire, cannula and tube were then dropped into a beaker receiving running hot water (about 80° C) and there allowed to remain for about five minutes. On removal from this the parts were shaken free from water, passed through 95 per cent alcohol and again shaken free, passed through ether and let dry.

By having a half-dozen cannulas and wires of standard size, it was possible to save trouble by cleaning a number at one time.

Not infrequently the first few samples of blood taken from an animal showed rapid or somewhat irregular rates of clotting. Some causes for these initial variations will be presented in following papers. The fairly uniform rate of clotting in any individual after the initial stage, varied in 21 different animals from an average of 3 to an average of 10.6 minutes, with a combined average of 5.9 minutes. The conditions for these variations among individuals have not been wholly determined.

The method here described has been employed not only to determine the coagulation time of cat's blood, but also that of human blood taken repeatedly through the skin at the base of the thumb nail. The skin was washed in soap and water, and rinsed in alcohol and ether before each test and then punctured with a sharp, well-pointed, three-cornered needle. The drop of blood that appeared on the skin was drawn into the cannula, and then the procedure was that above described. Following are figures obtained on one occasion:

Coagulation time	Variations from the average
5.5 minutes	+ 0.6 minutes
5 "	+ 0.1 "
4.5 "	− 0.4 "
4.5 "	− 0.4 "
5 "	+ 0.1 "
Average 4.9 minutes	Average error ± 0.3 minutes

Per cent average error = 6.

Doubtless if the lever were moved more frequently than every half-minute the average error would be reduced.

FACTORS AFFECTING THE COAGULATION TIME OF BLOOD

II. The Hastening or Retarding of Coagulation by Adrenalin Injections

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IN 1903, while tracing in dogs the course of adrenalin hyperglycaemia, Vosburgh and Richards first noted that simultaneously with the increase of blood sugar there occurred more rapid coagulation of the blood. In some cases the diminution was as much as four-fifths the coagulation time of the control. Since this result was obtained by painting adrenalin on the pancreas, as well as by intraperitoneal injection, they concluded that "the phenomenon appears to be due to the application of adrenalin to the pancreas."[1] Six years later during a study of the effect of adrenalin on internal hemorrhage, Wiggers examined incidentally the evidence presented by Vosburgh and Richards, and after many tests on five dogs found "never the slightest indication that adrenalin, either when injected or added to the blood, appreciably hastened the coagulation process."[2] In 1911, von den Velden reported that adrenalin (about 0.007 mg. per kilo) decreased the coagulation time in man about one-half — an effect appearing 11 minutes after administration by mouth, and 85 minutes after subcutaneous injection. He affirmed also, but without describing the conditions or giving figures, that adrenalin decreases coagulation time in vitro. He did not attribute the coagulative effect of adrenalin in patients to this direct action on the blood, however, but to vasoconstriction disturbing the normal circulation and

[1] Vosburgh and Richards: This journal, 1903, ix, p. 39.

[2] Wiggers: Archives of internal medicine, 1909, iii, p. 152.

thereby the normal equilibrium between blood and tissue. In consequence, the tissue juices with their coagulative properties enter the blood, so he assumes. In support of this theory he offers his observation that coagulation time is decreased after the nasal mucosa has been rendered anemic by adrenalin pledgets.[1] Von den Velden's claim for adrenalin given by mouth was subjected to a single test on man by Dale and Laidlaw, but their result was completely negative." [2]

The importance of Vosburgh and Richard's observation, the thoroughly discordant testimony of later investigators, as well as the meager and incidental nature of all the evidence that has been adduced either for or against the acceleration of clotting by adrenalin, made desirable a further study of this matter. In doing so we have employed cats as subjects. Usually they were quickly decerebrated under ether, and then continuance of the drug became unnecessary. Body temperature was maintained by means of an electric heating pad. Respiration proceeded normally except in a few instances (in which, presumably, there was hemorrhage into the medulla) when artificial respiration had to be given. The drawing of blood and the recording of coagulation were accomplished by methods already described.[3] The adrenalin used was that prepared by Parke, Davis and Co.; it was injected either subcutaneously or intravenously.

The effects of subcutaneous injections. — The first observations were of this class.

Oct. 27. A cat weighing about 3 k. was given 3 c.c. adrenalin 1: 1000, i.e., 1 mg. per kilo, under the skin. The animal, in this instance, was kept in uniform ether anaesthesia. Following is a record showing when blood was taken, and the coagulation time in each instance:

[1] Von den Velden: Münchener medizinische Wochenschrift, 1911, lviii, p. 187.

[2] Dale and Laidlaw: Journal of pathology and bacteriology, 1912, xvi, p. 362.

[3] Cannon and Mendenhall: This journal, 1914, xxxiv, p. 225.

2.56 — Injection made	3.27 — 3.5 minutes
.59 — 6 minutes	.44 — 2 "
3.07 — 5.5 "	.55 — 2.5 "
.13 — 5 "	4.07 — 3 "
.20 — 6.5 "	.20 — 2 "
Average 5.7 minutes	2.6 minutes

4.44 — 6 minutes
5.00 — 4.5 "
5.50 — 5 "
Average 5.2 minutes

In this case the coagulation time remained at its usual level for about 20 minutes after the subcutaneous injection.[1] Thereafter for about an hour the coagulation time averaged 45 per cent of its previous duration. And widely separated tests made during the following hour indicated that approximately the initial rate of clotting had been regained.

The rather long period (nearly 30 minutes), in the case just cited, between the injection and the first appearance of rapid clotting was not the rule. As the following figures show, the coagulation time may become shortened quite promptly after subcutaneous injection, —

Oct. 29.	3.30 — 5.5 minutes	4.01 — 3.5 minutes
	.36 — 5.5 "	.08 — 3.5 "
	.44 — 3 c.c. adrenalin (1: 1000) injected	.16 — 4.5 "
	.46 — 5.5 minutes	.23 — 5 "
	.53 — 4 "	.30 — 5.5 "

In this case nine minutes after the injection the change in the rate of clotting had begun, and it continued more rapid for the subsequent half-hour.

[1] This period is longer than is expected after the subcutaneous injection of any drug. As will be shown later, *strong* doses of adrenalin injected rapidly may not at first shorten the clotting process. Probably in some instances of subcutaneous injection of these strong doses, the drug enters the circulation more rapidly than in others and in consequence coagulation is not at first accelerated.

We did not attempt to find the minimal *subcutaneous* dose which would shorten clotting. A dose of 0.01 mg. per kilo, however, has proved effective, as shown by the following figures:

Feb. 3. 11.34 — 10 minutes
.45 — 9 "
.50 to .52 " Adrenalin, 2.8 c.c., 1: 100,000, injected under skin of groin in cat weighing 2.8 k.
.55 — 10 minutes
12.06 — 7 "
.14 — 4 "
.19 — 5.5 "
.31 — 6 "
.37 — 7 "
.45 — 9 "

As will be shown later, the dose in this instance was ten times the minimal effective *intravenous* dose. On the basis of these figures, less than a milligram of adrenalin given subcutaneously would be necessary to shorten clotting to a marked degree in a man of average weight (70 kg).

Not many observations were made by us on the effects of adrenalin administered subcutaneously. The amount of adrenalin reaching the vascular system and the rate of its entrance into the blood could be so much more accurately controlled by intravenous than by subcutaneous introduction that most of our attention was devoted to the latter method.

The effect of intravenous injections. — In this procedure a glass cannula was fastened in one of the external jugular veins, and filled with the same solution as that to be injected. A short rubber tube was attached and tightly clamped close to the glass. Later, for the injection, the syringe needle was inserted through the rubber and into the fluid in the cannula, the clip on the vein was removed, and the injection made.

The solutions employed intravenously were adrenalin 1: 10,000, 1:50,000, and 1:100,000 in distilled water.

The smallest amount which produced any change in clotting time was 0.1 c.c. of 1:100,000, in a cat weighting 2 kg., a dose of 0.0005 mg. per kilo. Four tests previous to the injection averaged 5 minutes, and none was shorter than 4 minutes. Immediately after the injection the time was 2 minutes, but at the next test the effect had disappeared. Doubling the dose in the same cat — i.e., giving 0.2 c.c. (0.001 mg. per kilo) — shortened the coagulation time for about 40 minutes:

Dec. 23.	10.30 — 4 minutes		11.00 — 1.5 minutes
	.35 — 4 "		.05 — 1.5 "
	.41 — 4 "		.10 — 3 "
	.46 —	Adrenalin, 0.001 mg. per k.	.15 — 2 "
	.47 — 2.5 minutes		.20 — 4 "
	.50 — 3.0 "		.26 — 4.5 "
	.55 — 3.5 "		.31 — 5 "

From 10.47, immediately after the second injection, till 11.20, the average time for clotting was 2.5 minutes, whereas both before and after this period the time was 4 minutes or longer. At 11.00 o'clock and 11.05, when the end point was reached in 1.5 minutes (a reduction of 63 per cent), a thick jelly was found on examining the cannula. The changes in clotting time in this case are represented graphically in Figure 1.

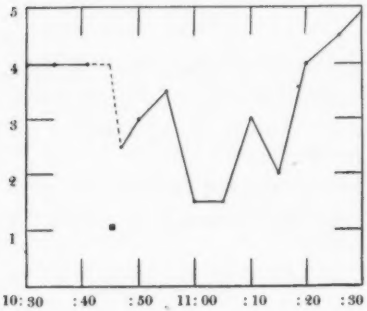

FIGURE 1. Shortening of coagulation time after injecting adrenalin, 0.2 c.c. 1: 100-000 (0.001 mg. per k.), at 10.46.

In another case a dose of 0.0005 mg. per kilo failed to produce any change, but 0.001 mg. per kilo (0.28 c.c. adrenalin 1:100,000, to a cat weighing 2.8 kg.) brought a sharp decline in the record, as follows:

Jan. 9.	11.32 — 6 minutes		11.55 — 4 minutes
	.40 — 6 "		12.00 — 5.5 "
	.47 —	Adrenalin, 0.001 mg. per k.	.06 — 7 "
	.48 — 5.5 minutes		

In these instances the animals were decerebrated. For decerebrate cats, the least amount of adrenalin, intravenously, needed to produce shortening of coagulation time is approximately 0.001 mg. per kilo.

In the above cases rapid clotting was manifest directly after minute doses. Larger doses, however, may produce primarily not faster clotting but slower, and that may be followed in turn by a much shorter coagulation time. The figures below present such an instance:

Nov. 25.	2.36 — 3 minutes		3.03 — 1.5 minutes
	.40 — 3 "		.05 — 1.5 "
	.43 —	Adrenalin, 0.5 c.c., 1: 10,000	.07 — 2.5 "
	.44 — 4 minutes		.10 — 1.5 "
	.49 — 3.5 "		.14 — 1.5 "
	.53 — 1.5 "		.16 — 2.5 "
	.55 — 1.5 "		.19 — 3 "
	.58 — 2 "		.23 — 3 "
	3.00 — 2.5 "		.30 — 3 "

This unexpected primary increase of coagulation time, lasting at least six minutes, is in striking contrast to the later remarkable shortening of the process from 3 to an average of 1.7 minutes for more than 20 minutes (see Figure 2, A).

If a strong solution, i.e., 1:10,000, is injected rapidly, the process may be prolonged as above, but not followed as above by shortening, thus:

Nov. 28.	9.59 — 3 minutes		10.18 — 3.5 minutes
	10.03 — 3 "		.22 — 3.5 "
	.08 —	Adrenalin, 0.5 c.c., 1: 10,000	.26 — 3 "
	.10 — 3 minutes		.29 — 3 "
	.14 — 3.5 "		.33 — 3 "

There was in this case no decrease in coagulation time at any test for a half-hour after the injection but instead a lengthening (see Figure 2, B). Howell has reported the interesting observation that repeated massive doses of adrenalin given to dogs may so

greatly retard coagulation that the animals may be said to be haemophilic.[1] These two instances show that on coagulation large doses have the contrary effect to small, just as has been shown to be true for intestinal and arterial smooth muscle.[2]

In a few experiments the brain and the cord to midthorax were destroyed through the orbit. Artificial respiration then maintained the animal in uniform condition. Under these circumstances, adrenalin intravenously had more lasting effects than when given to the usual decerebrate animals with intact cord. Figure 3 illustrates such a case. For 30 minutes before injection the clotting time averaged 5.4 minutes. Then, about 10 minutes after 1 c.c.

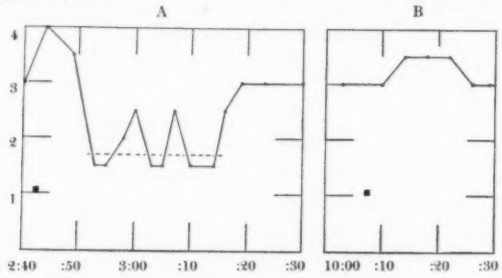

FIGURE 2. *A*, Primary lengthening followed by shortening of the coagulation time when adrenalin, 0.5 c.c. 1: 10,000 (0.05 mg.), was injected slowly, at 2.43. *B*, Lengthening of the coagulation time without shortening when the same dose was injected rapidly, at 10.08.

adrenalin 1:50,000 had been slowly injected, clotting began to quicken; during the next 20 minutes the average was 3.4 minutes, and during the following 45 minutes the average was 1.9 minutes — only 35 per cent as long as it had been before the injection.

In another case in which the brain and upper cord were similarly destroyed, the clotting time which for a half-hour had averaged 3.9 minutes was straightway reduced by 1 c.c. 1:100,000 to average for the next hour and 40 minutes 2.3 minutes, with 1.5 and 3 minutes as extremes. During the first 40 minutes of this period of 1 hour and 40 minutes of rapid clotting all of eight tests except two showed a coagulation time of 2 minutes or less.

[1] HOWELL: This journal, 1914, xxxiii, p. xiv.

[2] HOSKINS: This journal, 1912, xxix, p. 365. CANNON and LYMAN: This journal, 1913, xxxi, p. 376.

The explanation of this persistent rapid clotting in animals with spinal cord pithed is not yet clear.

As indicated in Figures 1, 2 and 3, the records of coagulation show oscillations. Some of these ups and downs are, of course, within the limits of error of the method, but in our experience they have occurred so characteristically after injection of adrenalin, and so often have appeared in a rough rhythm, that they have given the impression of being real accompaniments of faster clotting. It may be that two factors are operating, one tending to

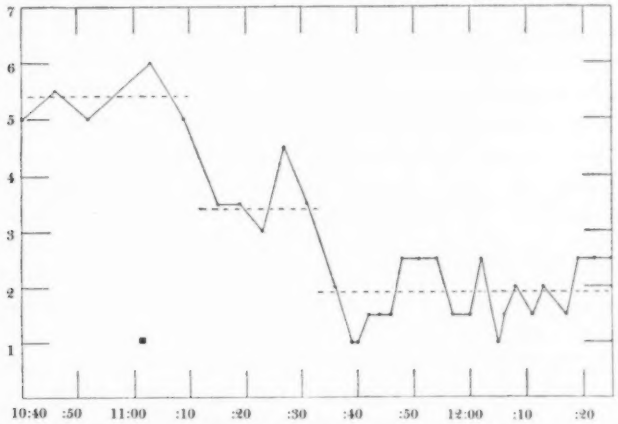

FIGURE 3. Prolonged shortening of the coagulation time after injecting (in an anima with brain and upper cord pithed) adrenalin 1 c.c. 1: 50,000 (0.02 mg.) at 11.01-02 The dash-lines represent averages.

hasten, the other to retard the process, and that the equilibrium disturbed by adrenalin is recovered only after interaction to and fro between the two factors.

The oscillations in coagulation time after adrenalin injections suggest that it might vary with changes in blood pressure, for that also commonly oscillates after a dose of adrenalin. Simultaneous recording of blood pressure and determining of coagulation time have revealed that each may vary without noteworthy variation in the other. Within ordinary limits, therefore, changes of blood pressure do not change the rate of clotting.

As previously stated, von den Velden has contended that the shortening of coagulation time by adrenalin is due to exudation of

tissue juices resulting from vasoconstriction. The least amount of adrenalin which produces markedly faster clotting in the cat, however, is 0.001 mg. per kilo. It has been shown that this amount when injected slowly, as in the present experiments, results in brief vasodilation rather than vasoconstriction.[1] Von den Velden's explanation can therefore not be applied to these experiments.

He has claimed, furthermore, that adrenalin added to blood in vitro makes it clot more rapidly, but, as already noted, he gives no account of the conditions of his experiments and no figures. It is impossible, therefore, to criticise them. His claim, however, is contrary to Wigger's earlier observations[2] that blood with added adrenalin coagulates no more quickly than blood with an equal amount of added physiological salt solution. Also contrary to this claim are the following two experiments. (1) Ligatures are tied around the aorta and inferior vena cava immediately above the diaphragm, and thus the circulation is confined almost completely to the anterior part of the animal. Indeed, since the posterior part ceases to function in the absence of blood supply, the preparation may be called an "anterior animal." When such a preparation is made and 0.5 c.c. adrenalin 1:100,000 (half the usual dose because, roughly, half an animal) is injected slowly into one of the jugulars, coagulation is not shortened. Whereas for a half-hour before the injection the clotting time averaged 4.6 minutes, for an hour thereafter the average was 5.3 minutes — a prolongation which may have been due, not to any influence of adrenalin, but to failure of the blood to circulate through the intestines and liver.[3] In another experiment after the gastro-intestinal canal and liver had been removed from the animal, the average time for coagulation during 25 minutes before injecting adrenalin (0.23 c.c. 1:100,000 in an animal weighing originally 2.3 kg.) was 5.5 minutes, and during 40 minutes after the injection it was 6.8 minutes, with no case shorter than 6 minutes. In the absence of circulation through the abdominal viscera,

[1] CANNON and LYMAN: *loc. cit.*, p. 381.

[2] WIGGERS: *loc. cit.*, p. 152.

[3] See PAWLOW: Archiv für Physiologie, 1887, p. 458. BOHR: Centralblatt für Physiologie, 1888, ii, p. 263. MEEK: This journal, 1912, xxx, p. 173.

therefore, adrenalin fails to shorten the clotting time. (2) The cannulas are filled with adrenalin, 1:1000, and emptied just before being introduced into the artery. The small amount of adrenalin left on the walls is thus automatically mixed with the drawn blood. Alternate obs vations with these cannulas wet by adrenalin and with the usual dry cannulas show no noteworthy distinction:

Feb. 19. 2.21 — 6.0 minutes, with usual cannula
.30 — 6.5 " " " "
.36 — 6.5 " " adrenalin cannula
.49 — 6.0 " " " "
.56 — 7.0 " " usual cannula
3.04 — 6.0 " " adrenalin cannula

The results of these experiments have made it impossible for us to concede either of von den Velden's claims, i.e., that clotting occurs faster because adrenalin is added to the blood or because adrenalin, by producing vasoconstriction, causes tissues to exude coagulant juices.

Vosburgh and Richards found that coagulation became more rapid as the blood sugar increased. Conceivably faster clotting might result from this higher percentage of blood sugar. Against this assumption, however, is the fact that clotting is greatly accelerated by 0.001 mg. per kilo, much less than the dose necessary to increase the sugar content of the blood.[1] And furthermore, when dextrose (3 c.c. of a 10 per cent solution) is added to the blood of an anterior animal, making the blood sugar roughly 0.3 per cent, the coagulation time is not markedly reduced. Adrenalin appears to act, therefore, in some other way than by increasing blood sugar.

Since adrenalin makes the blood clot much faster than normally in the intact animal, and fails to have this effect when the circulation is confined to the anterior animal, inference is justified that in the small doses here employed adrenalin produces its remarkable effects, not directly on the blood itself, nor through changes

[1] CANNON: This journal, 1914, xxxiii, p. 396.

in the extensive neuromuscular, bony, or surface tissues of the body, but through some organ in the abdomen.

That exclusion of the liver from the bodily economy by ligature of its vessels or by phosphorus poisoning,[1] will result in great lengthening of the coagulation time has been clearly shown. The liver, therefore, seems to furnish continuously to the blood a factor in the clotting process which is being continuously destroyed in the body. It is not unlikely that adrenalin makes the blood clot more rapidly by stimulating the liver to discharge this factor in greater abundance. But proof for this suggestion has not yet been established.

Summary

Adrenalin injected in small doses intravenously (0.001 mg. per kilo) and in larger doses subcutaneously, will shorten coagulation time to one-half or one-third the former duration.

The prompt shortening of the process after small doses is changed after larger doses (about 0.03 mg. per kilo) to a lengthening and later a shortening, or to a lengthening alone.

The effect of adrenalin on the clotting time is not associated with any corresponding effect on arterial pressure.

If the blood is confined anterior to the diaphragm, or if the intestines and liver are removed, adrenalin in small doses does not cause rapid clotting.

The addition of small amounts of adrenalin to drawn blood does not hasten clotting.

Increase of dextrose in the blood to 0.3 or 0.4 per cent does not cause the rapid clotting seen after adrenalin injection.

The explanation is suggested that adrenalin accelerates the clotting process by stimulating the liver (and intestines?) to greater activity in discharging some factor or factors in coagulation.

[1] See Meek: *loc. cit.*, p. 170. Also Whipple and Hurwitz: Journal of experimental medicine, 1911, xiii, p. 136.

FACTORS AFFECTING THE COAGULATION TIME OF BLOOD

III. The Hastening of Coagulation by Stimulating the Splanchnic Nerves

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IN a previous paper in this series evidence was presented that the intravenous injection of minute amounts of adrenalin hastens the clotting of blood.[1] The amounts used did not vary much above or below the amounts discharged by the adrenal glands after brief stimulation of the splanchnic nerves, as determined by Osgood in this laboratory,[2] and may therefore be regarded as physiological. Since injected adrenalin is capable of shortening the coagulation time, might not the increased secretion of the adrenals resulting from splanchnic stimulation likewise have that effect? The answer to that question was the object of the experiments here recorded.

The blood was taken and its coagulation was recorded graphically in the manner previously described.[3] In some instances the cats were etherized, in others they were anaesthetized with urethane, or were decerebrated. The splanchnic nerves always were stimulated after being cut away from connection with the spinal cord. Sometimes the nerves were isolated unilaterally in the abdomen; sometimes, in order to avoid manipulation of abdominal viscera, they were isolated in the thorax and stimulated singly or together. A tetanizing current was used, barely perceptible on the tongue and too weak to cause by spreading any contraction of skeletal muscles.

[1] Cannon and Gray: This journal, 1914, xxxiv, p. 235.

[2] See Cannon, This journal, 1914, xxxiii, p. 369.

[3] See Cannon and Mendenhall: This journal, 1914, xxxiv, p. 225.

The effects of splanchnic stimulation. — That splanchnic stimulation accelerates the clotting of blood, and that the effects vary in different animals, are facts illustrated in the following cases:

Oct. 25. A cat was etherized and maintained in uniform ether anaesthesia. After forty minutes of preliminary observation the left splanchnic nerves were stimulated in the abdomen. Following are the figures which show the effects on the coagulation time:

3.00 — 4 minutes	4.03 — 2.5 minutes
.07 — 5.5 "	.07 — 2.5 "
.14 — 4 "	.11 — 3 "
.32 — 4.5 "	.16 — 2 "
.39 to .40 Stim. left spl.	.20 — 1.5 "
.42 — 5 minutes	.23 — 4 "
.49 — 5 "	.29 — 5.5 "
.56 — 2 "	.40 — 5.5 "
4.00 — 1 "	.50 — 5 "

In this instance at least ten minutes elapsed between the end of stimulation and the beginning of faster clotting. The period of faster clotting, however, lasted for about a half-hour, during which the coagulation time averaged 2.1 minutes, only 43 per cent of the previous average of 4.8 minutes. It is noteworthy that the curve (see Figure 1), while lower, shows oscillations not unlike those which follow injection of adrenalin.[1]

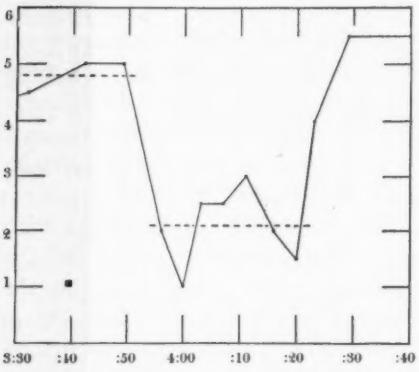

FIGURE 1. Shortening of coagulation time after stimulation of the left splanchnic nerves 3.39 — .40.

The primary delay of the effect is not always, indeed it is not commonly, present:

Nov. 6. A cat was anaesthetized (1.40 P.M.) with urethane, and later (3.05) its brain was pithed. The following observations on the

[1] Cf. CANNON and GRAY: This journal, 1914, xxxiv, p. 239.

coagulation time show the prompt effect of splanchnic stimulation:

3.36 — 7 minutes
.46 — 6 "
4.02 to .05 Stim. left spl. in abdomen
.08 — 4 minutes
.10 — 3 "
.18 — 3.5 "
.23 — 6.5 "

In Figure 2 is presented the original record of the shortening of coagulation after stimulation of the left splanchnic nerve (Nov. 8), in a cat with brain pithed.

In the foregoing instances the coagulation time was reduced after splanchnic stimulation to less than half what it was before. The reduction was not always so pronounced.[1]

Nov. 7. A cat maintained in uniform ether anaesthesia with artificial respiration had the following changes in the clotting time of its blood as the result of stimulating the left splanchnic nerve in the thorax:

3.40 — 5 minutes	4.11 — 4 minutes
.45 — 5 "	.16 — 3.5 "
.51 — 5.5 "	.21 — 4 "
.58 to 4.00 Stim. left spl.	.26 — 4.5 "
4.01 — 4.5 "	.31 — 5 "
.06 — 3.5 "	.36 — 6.5 "

In this case the average for about 15 minutes before stimulation was slightly over 5 minutes, and for 25 minutes thereafter it was four minutes.

[1] This animal had just passed through a period of excitement with rapid clotting (see CANNON and MENDENHALL: This journal, 1914, xxxiv, p.258.).

FIGURE 2. About two-fifths original size. Record of shortening of coagulation time after stimulation of the left splanchnic nerves, 4.33 — .35. The time before stimulation was 6 minutes, and afterwards 3, 4, 4, 4.5, and 6 minutes.

In all cases thus far the period of shortened coagulation lasted from 10 to 30 minutes. In other cases, however, the effect was seen only in a single observation. If this had occurred only once after splanchnic stimulation, it might be attributed to accident, but it was not an infrequent result, e.g.:

Oct. 28. A cat was etherized and decerebrated and the splanchnic nerves were isolated in the thorax. Following are two instances of brief shortening of coagulation after splanchnic stimulation:

3.36 — 4.5 minutes	4.07 — 4.5 minutes
.42 — 4.5 "	.12 — 5.5 "
.47 to .49 Spl. stim.	.19 to .22 Spl. stim.
.51 — 4.5 minutes	.23 — 3.5 minutes
.57 — 2 "	.27 — 4 "
4.01 — 4 "	.33 — 5 "

In the foregoing instance it is noteworthy that the degree of acceleration is not so great after the second stimulation of the splanchnics as it was after the first. This reduction of effect as the nerves were repeatedly stimulated was frequently noted. The following case presents another illustration:

Nov. 12. A cat was etherized (2.35 P.M.) and piqûre was performed (3.12) upon it. The operation was without effect. The loss or lessening of effectiveness on second stimulation of the left splanchnic nerves is to be compared with the persistence of effectiveness on the right side:

3.40 — 4.5 minutes	3.39 — 4 minutes
.45 — 4.5 "	.44 — 4 "
.54 to .56 Stim. left spl. in abd.	.48 — 4 "
4.00 — 3 minutes	.55 to .57 Stim. right spl.
.05 — 2 "	.59 — 3 minutes
.10 — 5.5 "	5.02 — 2.5 "
.16 — 5 "	.07 — 3 "
.22 to .27 Stim. left spl. in abd.	.11 — 3 "
.30 — 4 minutes	.15 — 5.5 "
.34 — 4 "	.22 — 5.5 "

The experiments above recorded show that stimulation of the splanchnic nerves results immediately, or after a brief period, in a

shortening of the coagulation time of the blood — an effect which in different animals varies in duration and intensity, and diminishes as the stimulation is repeated. The next question is whether this effect is produced through the adrenal glands.

The effect of splanchnic stimulation with and without the adrenal glands. — The manner in which splanchnic stimulation produces its effects is indicated in the following experiments:

Nov. 28. A cat was etherized and pithed through the orbit to the mid-thorax. The blood vessels of the *left* adrenal gland were then quickly tied and the gland removed. The readings for a half hour before the left splanchnic nerve was stimulated averaged 7 minutes, then, —

4.38 to 40 Stim. left spl. (glandless)
.42 — 7 minutes
.50 — 7 "
5.02 to .04 Stim. right spl.
.06 — 4 minutes
.10 — 7 "
.18 — 7 "
.26 — 7 "

Dec. 4. A cat was etherized and pithed through the orbit to the neck region. The right and left splanchnic nerves were tied and cut in the thorax. The *left* adrenal gland was then carefully removed. These operations consumed about a half-hour. The following records show the effect of stimulating the left and right splanchnic nerves:

4.10 — 5 minutes
.16 — 4.5 "
.25 to .28 Stim. left spl. (glandless)
.30 — 4.5 minutes
.35 — 4.5 "
.40 — 7.5 "
.49 to .51 Stim. right spl.
.55 — 4.5 minutes
5.00 — 2.5 "
5.14 — 6 minutes
.23 to .25 Stim. right spl.
.26 — 6 minutes
.33 — 4.5 "
.38 — 3.5 "
.43 — 4.5 "
.49 — 5 "
.55 — 6 "

The results in this experiment are represented graphically in Figure 3.

Elliott has presented evidence that in the cat the splanchnic innervation of the adrenals is not crossed, so that if the gland is removed on one side stimulation of the nerves on that side causes no discharge from the opposite gland.[1] As the above experiments clearly show, splanchnic stimulation on the glandless side results in no shortening of the coagulation time; whereas, in the same

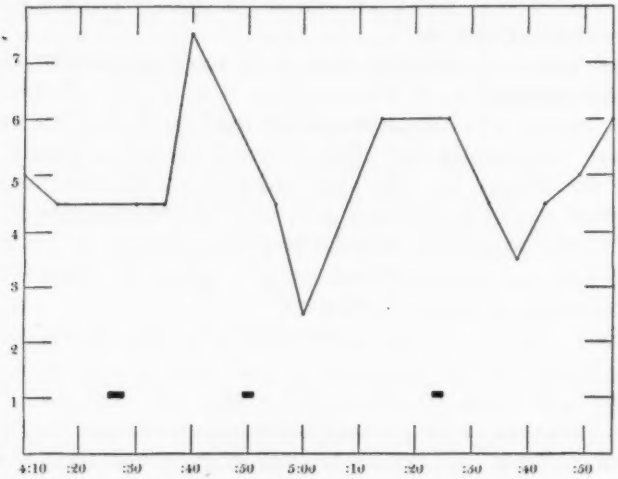

FIGURE 3. Results of stimulating the left splanchnic nerves, 4.25 — 28, after removal of the left adrenal gland, and of stimulating the right splanchnic nerves, 4.49 — .51 and 5.23 — .25, with right adrenal gland present.

animals, stimulation of the nerves on the other side (still connected with the adrenal gland) produces a sharp hastening of the clotting process.

The splanchnics innervate the intestines and liver even though the adrenal gland is removed. The foregoing experiments indicate that the nerve impulses delivered to these organs do not influence them in any direct manner to accelerate the speed of coagulation. Indeed in one of the experiments (Dec. 4, p. 247) a high reading about ten minutes after splanchnic stimulation on the glandless side suggests the possibility of an opposite effect. Direct stimulation

[1] ELLIOTT: Journal of physiology, 1912, xliv, p. 405.

of the hepatic nerves on one occasion was followed by a change of the clotting time from 4.5, 5, 4.5, 4.5 minutes during 25 minutes before stimulation to 4.5, 7, and 6 minutes during 20 minutes after stimulation.

Since with the adrenals present stimulation of hepatic nerves induces glycogenolysis and quick increase of blood sugar,[1] just as splanchnic stimulation does, the failure of the blood to clot faster after stimulation of the hepatic nerves confirms the evidence already offered that faster clotting in hyperadrenalinaemia is not due to hyperglycaemia.[2]

The liver and intestines cannot be made to shorten clotting time by stimulation of their nerves, but, as has already been shown, neither can adrenalin act by itself to hasten the clotting process.[3] Apparently the effect is produced by coöperation between the adrenals and the liver (and possibly also the intestines). Somewhat similar cooperation is noted in the organization of sugar metabolism; splanchnic stimulation in the absence of the adrenal glands does not increase blood sugar,[4] and in the absence of the liver adrenalin is without influence.[5]

The variations of effect noted after splanchnic stimulation can be accounted for by variations in the adrenalin content of the glands. Elliott reports that animals newly brought into strange surroundings may have a considerably reduced amount of adrenalin in their adrenals.[6] The animals used in our experiments had been for varying lengths of time in an animal house in which barking dogs were also kept, and were therefore subject to influences which would be likely to discharge the glands.

[1] MACLEOD: Diabetes: its Pathological Physiology, London, 1913, pp. 68–72.

[2] See CANNON and GRAY: This journal, 1914, xxxiv, p. 241.

[3] See CANNON and GRAY: *loc. cit.*, p. 240.

[4] GAUTRELET and THOMAS: Comptes rendus de la Société de Biologie, 1909, lxvii, p. 233.

[5] BANG: Der Blutzucker, Wiesbaden, 1913, p. 87.

[6] ELLIOTT: *loc. cit.*, p. 379.

SUMMARY

Stimulation of the splanchnic nerves results immediately, or after a brief delay, in shortening of the coagulation time of blood. The degree and the duration of the effect varies, — clotting not uncommonly takes less than half the time it took before stimulation, and this period of rapid clotting may last from 10 to 30 minutes.

The stimulation usually produces less marked effects as it is repeated.

If the adrenal gland is removed on one side, splanchnic stimulation on that side does not shorten the clotting time; whereas splanchnic stimulation on the other side is still effective. The faster clotting is therefore due to increased adrenal discharge.

Since stimulation of nerves supplying the liver and intestines does not hasten clotting, and since increase of adrenalin has no effect in the absence of the liver and intestines, the shortened clotting after splanchnic stimulation is accounted for by the action of adrenal discharge on the liver (and intestines?).

The variations in the effects in different animals can be accounted for by variations in the adrenalin content of the adrenal glands in confined animals.

FACTORS AFFECTING THE COAGULATION TIME OF BLOOD

IV. The Hastening of Coagulation in Pain and Emotional Excitement

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IN the preceding paper of this series evidence was given to prove that stimulation of splanchnic nerves, with accompanying increase of adrenal secretion, results in more rapid clotting of blood. Recent experiments have shown that certain conditions — such as pain and emotional excitement — likely to arise in the natural life of organisms and known to be attended by nervous discharges over splanchnic courses, are also attended by increased secretion of adrenalin into the blood.[1] Does the adrenalin thus liberated have any effect on the rate of coagulation? The observations here recorded were made in order to obtain an answer to that question.

The effect of "painful" stimulation. — In experiments on the action of stimuli which in the unanaesthetized animal would cause pain, faradic stimulation of a large nerve trunk (the stump of the cut sciatic), and operation under light anaesthesia, were the methods used to affect the afferent nerves. Elliott found that repeated excitation of the sciatic nerve was especially efficient in exhausting the adrenal glands of their adrenalin content, and also that this reflex persisted after removal of the cerebral hemispheres.[2] It was to be expected, therefore, that with well-stored glands, sciatic stimulation, even in the decerebrate animal, would call forth an amount of adrenal secretion which would decidedly hasten clotting. The following case illustrates such a result:

[1] Cannon: This journal, 1914, xxxiii, p. 357.

[2] Elliott: Journal of physiology, 1912, xliv, pp. 406, 407.

Dec. 12. A cat was anaesthetized with ether at 3.45, and the left sciatic nerve was bared. Decerebration was completed at 3.57. The clotting time of the blood began to be tested six minutes later:

4.03 — 4 minutes	4.45 to .50 Stim. left sciatic
.08 — 3.5 "	.53 — 2.5 minutes
.13 — 3.5 "	.57 — 7 "
.18 — 4.5 "	5.06 — 7.5 "
.23 to .25 Stim. left sciatic	.15 to .17 Stim. left sciatic
.26 — 2.5 minutes	.17 — 4 minutes
.29 — 3.5 "	.22 — 4.5 "
.34 — 4.0 "	.27 — 5.5 "
.40 — 5.0 "	.36 — 5.5 "
	.46 — 7 "

The results obtained in this case, which were similar to results in other cases, are represented graphically in Figure 1. The

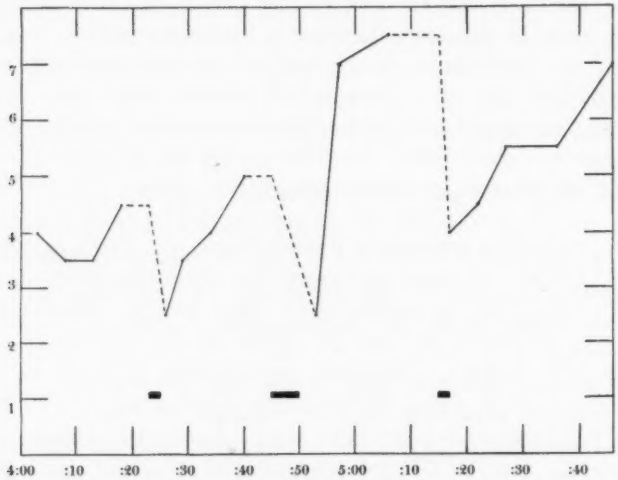

FIGURE 1. Three shortenings of coagulation time after stimulation of the left sciatic nerve, at 4.23 — .25, at 4.45 — 50 (stronger), and at 5.15 — .17.

coagulation time was becoming gradually more prolonged, but each excitation of the sciatic nerve was followed by a marked shorten-

ing. The strength of stimulation was not determined with exactness, but it is worthy of note that the current used in first and third stimulations was weaker than could be felt on the tongue, whereas that used in the second was considerably stronger, though it did not produce reflex spasms.

Mere tying of the nerve is capable of producing a marked shortening of coagulation, as the following figures show:

Oct. 21. 10.57 Cat under ether and urethane given
11.11 — 8.5 minutes
.23 — 8.5 "
.32 to .35 Left sciatic bared and tied
.37 — 1.5 minutes
.41 — 5.5 "
.50 — 7 "
12.02 — 8.5

Stimulation of the crural nerve had similar effects, reducing the clotting time in one instance from a succession of 3, 3, and 3.5 minutes to 1.5 minutes shortly after the application of the current, with a return to 3.5 minutes at the next test.

Operative procedures performed under light anaesthesia, or reduction of anaesthesia soon after operation, resulted in a remarkable shortening of the coagulation time:

Nov. 8. A cat was etherized and tracheotomized. The abdomen was then opened, and a ligature was drawn around the hepatic nerves. The operation was completed at 2.25. At 2.50 the etherization became light and the rate of clotting began to be faster:

2.50 — 6 minutes	.15 — 3.5 minutes
3.00 — 5.5 "	.20 — 4.5 "
.10 — 3.5 "	.30 — 7.5 "

Nov. 11. A female cat, very quiet, was placed in the holder at 1.55. The animal was not excited. At 2.10 etherization was begun; the animal was then tracheotomized, and the femoral artery was exposed.

2.21 — 4.5 minutes

.26 — 4.5 " Anaesthesia lessened

.32 — 3.5 " " light

.35 — Abdomen opened

.47 — 1.5 minutes

.52 — 1 "

.55 — Ligature passed around hepatic nerves

.57 — 1.5 minutes Anaesthesia light; corneal reflex present

3.02 — 3 "

.07 — 3 " Some hepatic nerves cut

.12 — 4.5 " Rest of hepatic nerves cut

.22 — 5 "

The results of this experiment are shown graphically in Figure 2.

Nov. 13. A cat was etherized at 1.55, tracheotomized, and the femoral artery laid bare. As soon as these preparations were completed, the ether was removed and anaesthesia became light. The blood clotted thus:

2.08 — 6 minutes,

.15 — 4 " Anaesthesia light

.20 — 2 "

.24 — 1 " Etherization begun again

.27 — 2.5 "

.30 — 3.5 "

.35 — 5.5 "

.50 — 5.5 "

In the foregoing and in other similar instances, a condition of surgical injury, whether just made or being made, was accompanied by more rapid clotting of blood when the degree of anaesthesia was lessened. This condition was one which, if allowed to go further in the same direction, would result in pain. Both direct electrical stimulation and also surgical operation of a nature to

give pain in the unanaesthetized animal, result, therefore, in faster clotting. It is worthy of note that after decerebration clotting apparently occurred no faster because the abdomen had been opened, although in the decerebrate state etherization was suspended. The mechanism for reflex control of the adrenals may not be higher than the corpora quadrigemina, as Elliott has shown, but the discharge from the glands seems to be more certain to occur when the cerebrum is present and is permitted even slightly to operate.

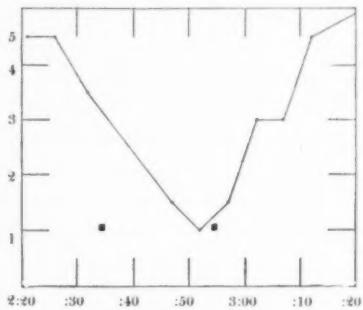

FIGURE 2. Shortening of coagulation time during an operation under light anaesthesia. At 2.35 the abdomen was opened, at 2.55 a ligature was passed around the hepatic nerves.

The effect of emotional excitement. — Reference has already been made to the emotional secretion of the adrenal glands. In their emotional reaction to being bound cats differ widely: some, especially young males, become furious; others, especially elderly females, take the experience quite calmly. This difference of attitude was used with positive results in experiments on emotional glycosuria,[1] and it seemed possible, therefore, to use it to test the effect of emotions on blood-clotting. To plan formal experiments for that purpose was not necessary, because in the ordinary course of the researches here reported, the difference in effects on the blood between the violent rage of vigorous young males and the quiet complacency of old females was early noted. Indeed the rapid clotting which accompanied excitement not infrequently made necessary an annoying wait till slower clotting would permit the use of experimental methods for shortening the process.

The animals used on November 11 and 13 (see pp. 253, 254) are examples of calm acceptance of being placed on the holder, and furthermore these animals were anaesthetized without much disturbance. As the figures indicate, from the first the clotting occurred at about the average rate.

[1] See CANNON, SHOHL, and WRIGHT: This journal, 1911, xxix, p. 280.

In sharp contrast to these figures are those obtained when a vigorous animal is angered:

Oct. 30. A very vigorous cat was placed on the holder at 9.08. It at once became stormy, snarling, hissing, biting, and lashing its big tail. At 9.12 etherizing was begun and that intensified the excitement. By 9.15 the femoral artery was tied. The clotting time of the blood for an hour after the ether was first given was as follows:

9.18 — 0.5 minute	9.43 — 1.0 minute
.19 — 1.0 "	.45 — 0.5 "
.22 — 1.0 "	.49 — 0.5 "
.24 — 1.0 "	.52 — 0.5 "
.26 — 1.0 "	.54 — 0.5 "
.28 — 1.5 "	.57 — 1.0 "
.31 — 1.0 "	10.00 — 0.5 "
.33 — 0.5 "	.02 — 0.5 "
.35 — 0.5 "	.06 — 1.0 "
.38 — 0.5 "	.09 — 0.5 "
.39 — 0.5 "	.11 — 0.5 "
.41 — 1.0 "	.13 — 1.0 "

Twenty-four observations made during the hour showed that the clotting time in this enraged animal averaged three-fourths of a minute and was never longer than a minute and a half. The clots were invariably a solid jelly. The persistence of the rapid clotting for so long a period after anaesthesia was started, may have been in part due to continued, rather light etherization, for Elliott found that etherization itself could reduce the adrenalin content of the adrenal glands.[1]

The shortened clotting did not always persist so long as in the foregoing instance. The brief period of faster clotting illustrated in the following case was typical of many:

Nov. 18. A cat that had been in stock for some time was placed on the holder at 2.13, and was at once enraged. Two minutes later

[1] ELLIOTT: *loc. cit.*, p. 388.

etherization was started. The hairs on the tail were erect. The clotting was as follows:

2.25 — 1.0 minute	.31 — 4.5 minutes
.27 — 0.5 "	.37 — 3.5 "
.28 — 2.0 "	.47 — 4.5 "

It seems probable that in this case, because of the cat's being caged near dogs, the adrenals were well-nigh exhausted previous to this experiment, and that the emotional flare-up practically discharged the glands, for repeated attempts later to reproduce the initial rapid clotting by stimulation of the splanchnic nerves was without result.

Evidence presented in previous papers of this series makes wholly probable the correctness of the inference that the faster coagulation which follows emotional excitement is due to adrenal discharge from splanchnic stimulation. In this relation the effect of severance of the splanchnics on emotional acceleration of the clotting process is of interest. The following cases are illustrative:

Oct. 29. A cat was left on the holder for 10 minutes while the femoral artery was uncovered under local anaesthesia. The blood removed was clotted in a half-minute. The animal was much excited. It was now quickly etherized and the brain pithed forward from the neck. The tests resulted as follows:

10.51 — 1.0 minute	
.53 — 05. "	
.55 — 0.5 "	
.57 — 0.5 "	
11.07 —	Cut left splanchnic
.12 —	" right splanchnic
.21 — 3.5 minutes	
.26 — 3.5 "	

The original record of this case is given in Figure 3.

Nov. 5. A cat was etherized at 2.35. At 2.39 artificial respiration by tracheal cannula was begun, the air passing through an ether bottle. The clotting occurred thus:

2.53 — 1.5 minutes
.57 — 1.5 "
3.05 — 1.5 "
.15 — 1.5 "
.25 — Both spl. cut and tied in thorax
.35 — 4.5 minutes
.55 — 4.5 "

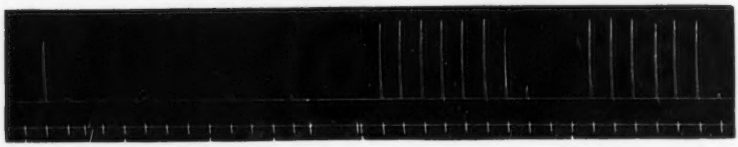

FIGURE 3. About two-thirds original size. Record of rapid clotting (less than a half-minute) after emotional excitement. At 11.07 the left, at 11.12 the right splanchnic nerves were cut; the clotting then required 3.5 minutes. The marks below the time record indicate the moments when the samples were drawn.

Nov. 7. A cat was etherized under excitement and with tail-hairs erect, at 1.55. At 2.13 the animal was showing reflexes. The figures show the course of the experiment:

2.15 — 1.5 minutes
.21 — 1.0 "
.26 — 1.0 "
.31 — 1.0 "
.36 — 1.0 "
.41 — 1.0 "
.46 — 2.0 "
.51 — 2.0 "
3.06 — 2.0 minutes
.11 — 2.5 "
.26 — Cut left spl. in thorax
.35 — " right spl. in thorax
.40 — 5.0 minutes
.45 — 5.0 "
.51 — 5.5 "

In this instance the subsequent stimulation of the splanchnic nerves resulted again in faster clotting — a reduction from 5.5 minutes to 3.5 minutes.[1] The results from this experiment are shown graphically in Figure 4.

[1] See CANNON and MENDENHALL: This journal, 1914, xxxiv, p. 245, experiment on Nov. 7.

Discussion

The data presented in this paper show that such stimulation as in the unanaesthetized animal would cause pain, and also such emotions as fear and rage, are capable of greatly shortening the coagulation time of blood. These results are quite in harmony with the evidence previously offered that injected adrenalin and

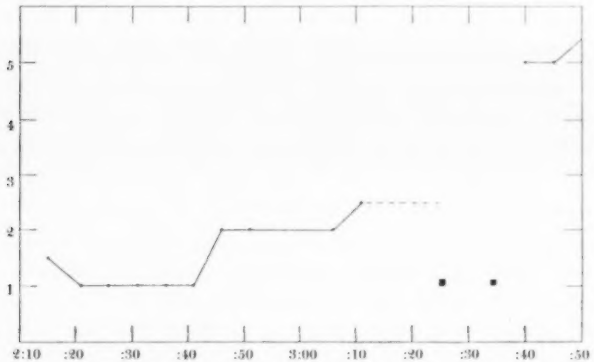

Figure 4. Rapid clotting after emotional excitement, with slowing of the process when the splanchnic nerves were cut in thorax (the left at 3.26, the right at 3.35).

secretion from the adrenal glands induced by splanchnic stimulation hasten clotting, for, as already stated, painful stimulation and emotional excitement also evoke activity of the adrenals.

In a previous paper the increase of blood sugar and the secretion of adrenalin in pain and the major emotions were interpreted as biological adaptations to conditions likely to involve, in wild life, pain and great emotion — i.e., the necessities of struggle, fighting or flight. The sugar would then serve the muscular energies, the adrenalin would aid in distributing blood to organs critically involved in the struggle, and would also abolish or minimize the effects of fatigue.[1] The more rapid clotting of blood in pain and emotional excitement may also be regarded as an adaptive process, useful to the organism. The importance of conserving the blood needs no argument. The effect of local injury in favoring the formation of a clot to seal the opened vessels

[1] See Cannon: This journal, 1914, xxxiii, p. 372.

is obviously adaptive in protecting the organism against hemorrhage. The injury that causes opening of blood vessels, however, is, if extensive, likely also to produce pain. And, as shown above, conditions producing pain increase adrenal secretion and hasten coagulation. Thus injury would be made less dangerous as an occasion for serious hemorrhage by two effects which the injury itself produces in the body, — the local effect on clotting at the region of injury and the general effect on the speed of clotting wrought by reflex secretion of adrenalin.[1]

The strong emotions, as fear and anger, are reasonably regarded as the concomitants of bodily changes which may be of utmost service in subsequent action. These bodily changes are so much like those which occur in pain and fierce struggle that, as early writers on evolution suggested, the emotions may be considered as foreshadowing the suffering and intensity of actual strife. On this general basis, therefore, the bodily alterations attending violent emotional states would, as organic preparations for fighting and possible injury, naturally involve the effects which pain itself would produce. And rapid clotting, like increased blood sugar, increased adrenalin, and an adapted circulation, would be favorable to the preservation of the organism that could best effect it.[2]

[1] The conditions under which pain and danger from hemorrhage occur together in civilization are not so frequent as to permit a ready testing on man of the ideas here propounded. It is possible that the pain of childbirth is such as to lead to adrenal discharge (the increase of blood-sugar during parturition is indicative of that), and thus to favor rapid clotting at a time when that may be important.

[2] There is evidence that asphyxia causes increased secretion of adrenalin (see CANNON: This journal, 1914, xxxiii, p. 357), and often in the course of these investigations we have noted that when respiration became impaired and the blood turned dark, clotting was faster. In a few observations, however, in which a formal attempt was made to determine the influence of asphyxia on clotting, the results were not always positive — due probably to the use of animals which had been kept for some time in conditions likely to discharge the adrenal glands.

Stewart (Journal of experimental medicine, 1912, xv, p. 547) and Hoskins and McPeek (Journal of the American Medical Association, 1913, lx, p. 1778) have reported that direct massage of the adrenal glands causes an increased secretion of adrenalin. The increase, according to the latter authors, is slight. Mr. Horace Gray and Mr. C. A. L. Binger have tried in this laboratory the

SUMMARY

Stimulation of afferent nerves (sciatic, crural), or major operations under light anaesthesia, markedly shorten the coagulation time of blood.

Emotional excitement is the occasion for very rapid clotting (sometimes in less than a half-minute), which becomes slow (three to five minutes) when the splanchnic nerves are cut.

Pain and strong emotions have been proved to evoke secretion of the adrenal glands; and adrenalin hastens clotting. Rapid coagulation may reasonably be considered, therefore, as another instance of adaptive reaction serviceable to the organism in the injury which may accompany pain or which may follow the struggle that fear or rage may occasion.

possibility of influencing the coagulation of blood by massage of the adrenals. In one instance the clotting time was 5.5 minutes in four successive tests before massage. After massage it fell to an average of 4.6, for forty minutes. Then the glands were manipulated again, and the eight tests taken thereafter were as follows: 3.5, 4.5, 4, 4, 3.5, 4.5, 3.5, 4. In another instance massage of the glands reduced the time, after a brief prolongation, from an average of 6.1 minutes to an average of 4.7 minutes, with six tests in each group. In other cases the tests were not so favorable. Massage of the liver was without clear effect.